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=> s monooxygenase and oxidation
 L1 7515 MONOOXYGENASE AND OXIDATION

=> dup rem l1
 PROCESSING IS APPROXIMATELY 19% COMPLETE FOR L1
 PROCESSING IS APPROXIMATELY 52% COMPLETE FOR L1
 PROCESSING IS APPROXIMATELY 72% COMPLETE FOR L1
 PROCESSING IS APPROXIMATELY 95% COMPLETE FOR L1
 PROCESSING COMPLETED FOR L1
 L2 4386 DUP REM L1 (3129 DUPLICATES REMOVED)

=> s l2 and cytochrome P-450cam
 4 FILES SEARCHED...
 L3 28 L2 AND CYTOCHROME P-450CAM

=> s l3 and holoxygenated aromatic
 L4 0 L3 AND HOLOGENATED AROMATIC

=> s l3 and halogenated aromatic
 L5 0 L3 AND HALOGENATED AROMATIC

=> s l3 and halo aromatic
 L6 0 L3 AND HALO AROMATIC

=> s l3 and aromatic
 L7 5 L3 AND AROMATIC

=> d l7 1-5 ibib ab

L7 ANSWER 1 OF 5 MEDLINE
 ACCESSION NUMBER: 97163846 MEDLINE
 DOCUMENT NUMBER: 97163846 PubMed ID: 9010601
 TITLE: A structure-based model for cytochrome P450cam-
 putidaredoxin interactions.
 AUTHOR: Pochapsky T C; Lyons T A; Kazanis S; Arakaki T; Ratnaswamy
 G
 CORPORATE SOURCE: Department of Chemistry, Brandeis University, Waltham, MA
 02254-9110, USA.
 CONTRACT NUMBER: R01-GM-44191 (NIGMS)

SOURCE: BIOCHIMIE, (1996) 78 (8-9) 723-33.
 Journal code: 1264604. ISSN: 0300-9084.
 PUB. COUNTRY: France
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 199707
 ENTRY DATE: Entered STN: 19970721
 Last Updated on STN: 19970721
 Entered Medline: 19970708

AB Putidaredoxin (Pdx) is a Fe₂S₂ ferredoxin which acts as the physiological reductant of **cytochrome P-450cam** (CYP101). A model for the solution structure of oxidized Pdx has been determined using NMR methods (Pochapsky et al (1994) Biochemistry 33, 6424-6432). ¹H-¹⁵N correlations and redox-dependent amide exchange rates have also been described (Lyons et al (1996) Protein Sci 5, 627-639). Data obtained from mutagenesis and kinetic measurements concerning the interactions of Pdx and CYP101 are summarized. A model for the structure of the homologous ferredoxin adrenodoxin (Adx) is also described, and data concerning Adx activity are discussed in relation to this structure. The structures of Pdx and CYP101 were used as starting points for molecular modeling and molecular dynamics simulations. Close approach between the metal centers of the two proteins and interaction between **aromatic** residues on the surfaces of the proteins are premised. The resulting complex exhibits three intermolecular salt bridges, five intermolecular hydrogen bonds and a 12 Å distance between the metal centers. The first direct observations of interaction between Pdx and CYP101 (by two-dimensional NMR of ¹⁵N-labeled Pdx in solution with CYP101) are described. The results of the NMR experiments indicate that conformational gating of the electron transfer complex between CYP101 and Pdx may be important.

L7 ANSWER 2 OF 5 CAPLUS COPYRIGHT 2003 ACS
 ACCESSION NUMBER: 2002:220824 CAPLUS
 DOCUMENT NUMBER: 136:259213
 TITLE: Screening method for oxygenase enzymes using **aromatic** substrates which are converted to spectrochemically detected polymeric oxygenated compounds by a coupling enzyme
 INVENTOR(S): Arnold, Frances H.; Joo, Hyun
 PATENT ASSIGNEE(S): California Institute of Technology, USA
 SOURCE: PCT Int. Appl., 125 pp.
 CODEN: PIXXD2
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2002022861	A1	20020321	WO 2000-US28768	20001013
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG				
AU 2000080285	A5	20020326	AU 2000-80285	20001013
PRIORITY APPLN. INFO.:			US 2000-661093	A1 20000913
			WO 2000-US28768	W 20001013
AB A method is provided for detecting the presence of an oxygenated compd.				

which is produced when a substrate is reacted with an oxygenase for the substrate. The method involves reacting a coupling enzyme with the oxygenated compd. to form a polymeric oxygenated compd. which is fluorescent or luminescent. Measurement of the fluorescence or luminescence of the polymeric oxygenated compd. provides indirect detection of the oxygenated compd. produced by reaction of the oxygenase with the substrate. The method is carried out in a whole cell environment wherein the cell is transformed to express both the oxygenase being screened and the coupling enzyme. The method can be used to measure the activity of monooxygenases and dioxygenases on arom. substrates. Thus, for example, the activity of **cytochrome P 450cam** in *Escherichia coli* is checked by measuring the conversion of naphthalene to a hydroxylated product (e.g., 1-naphthol, 2-naphthol) which emits a blue fluorescence when exogenously added horseradish peroxidase polymerizes the product. The method is amenable to large scale screening of enzyme mutants to isolate those with max. oxygenase activity. Thus, a screening strategy with high throughput fluorescence image anal. was implemented in order to identify bacterial clones expressing improved hydroxylating enzymes. Mutants of P 450cam with improved activity on naphthalene (or 3-phenylpropionate) and H₂O₂ are identified.

REFERENCE COUNT: 2 THERE ARE 2 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 3 OF 5 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2000:911441 CAPLUS

DOCUMENT NUMBER: 134:68048

TITLE: Analogs of a cytochrome P450 of *Pseudomonas putida* with improved catalytic action **aromatic** halohydrocarbons for use in bioremediation of soil

INVENTOR(S): Wong, Luet Lok; Jones, Jonathan Peter

PATENT ASSIGNEE(S): Isis Innovation Limited, UK

SOURCE: PCT Int. Appl., 42 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: English

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2000078973	A1	20001228	WO 2000-GB2379	20000619
W:	AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM			
RW:	GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG			
EP 1190067	A1	20020327	EP 2000-942200	20000619
R:	AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO			

PRIORITY APPLN. INFO.: GB 1999-14373 A 19990618

WO 2000-GB2379 W 20000619

AB Analogs of **cytochrome P 450cam** of

Pseudomonas putida that have improved catalytic activity against heavily halogenated arom. hydrocarbons and that may be of use in the reclamation of soils contaminated with polychlorinated biphenyls. In particular, alterations in the substrate pocket that increase the vol. available for bulky polyhalogenated aroms. are described. Prepn. of a series of analogs of the gene camC cytochrome P 450 with increased activity towards polychlorinated biphenyls is demonstrated. A fusion protein of

putidaredoxin and putidaredoxin reductase that can be used as a cofactor is also described.

REFERENCE COUNT: 7 THERE ARE 7 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L7 ANSWER 4 OF 5 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 1994:133100 BIOSIS
DOCUMENT NUMBER: PREV199497146100
TITLE: Putative functions of phenylalanine-350 of Pseudomonas putida cytochrome P-450-cam.
AUTHOR(S): Yasukochi, Takanori; Okada, Osamu; Hara, Takayuki; Sagara, Yasuhiro (1); Sekimizu, Kazuhisa; Horiuchi, Tadao
CORPORATE SOURCE: (1) Dep. Med. Biol., Kochi Med. Sch., Okoh-cho, Nankoku, Kochi 783 Japan
SOURCE: Biochimica et Biophysica Acta, (1994) Vol. 1204, No. 1, pp. 84-90.
ISSN: 0006-3002.
DOCUMENT TYPE: Article
LANGUAGE: English

AB Cytochrome P-450-cam hydroxylates d-camphor, using molecular oxygen and reducing equivalents transferred via putidaredoxin. We constructed mutant genes in which Phe-350 of P-450-cam was replaced by Leu, Tyr, or His by site-directed mutagenesis, expressed them in Escherichia coli, purified the mutant proteins, and compared their enzymic properties with those of the wild type P-450-cam. NADH **oxidation** rate of the Tyr mutant in the reconstituted system with putidaredoxin and putidaredoxin reductase was similar to that of the wild type enzyme, while the Leu mutant and the His mutant showed 67% and 17% activity of that of the wild type, respectively. The affinities of these mutant proteins for camphor and the oxidized form of putidaredoxin were much the same as those of the wild type protein. Rate constants for the reduction reaction of P-450-cam by reduced putidaredoxin, a physiological electron donor for P-450-cam, of Tyr and His mutants were much the same as that of the wild type enzyme, whereas the Leu mutant showed approx. half that of the wild type. Thus, the **aromatic** ring of Phe-350 of P-450-cam probably contributes to enhancing efficiency of the electron transfer yet does not seem to be essential for the reaction.

L7 ANSWER 5 OF 5 SCISEARCH COPYRIGHT 2003 ISI (R)
ACCESSION NUMBER: 97:87565 SCISEARCH
THE GENUINE ARTICLE: WD134
TITLE: Gene organization and low regiospecificity in **aromatic**-ring hydroxylation of a benzene **monooxygenase** of Pseudomonas aeruginosa J1104
AUTHOR: Kitayama A (Reprint); Suzuki E; Kawakami Y; Nagamune T
CORPORATE SOURCE: UNIV TOKYO, GRAD SCH ENGN, DEPT CHEM & BIOTECHNOL, BUNKYO KU, 7-3-1 HONGO, TOKYO 113, JAPAN (Reprint); INST RES & INNOVAT, DEPT BIOTECHNOL, KASHIWA, CHIBA 277, JAPAN
COUNTRY OF AUTHOR: JAPAN
SOURCE: JOURNAL OF FERMENTATION AND BIOENGINEERING, (20 JAN 1996) Vol. 82, No. 5, pp. 421-425.
Publisher: SOC FERMENTATION BIOENGINEERING, JAPAN, OSAKA UNIV, FACULTY ENGINEERING, 2-1 YAMADAOKA, SUITA, OSAKA 565, JAPAN.
ISSN: 0922-338X.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE; AGRI
LANGUAGE: English
REFERENCE COUNT: 20

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB A novel benzene **monooxygenase** gene cluster has been cloned from Pseudomonas aeruginosa J104 which was isolated from soil as a benzene degrader. The nucleotide sequence of this gene cluster was found to be

highly homologous to those of other toluene **monooxygenase** gene clusters. This multicomponent **monooxygenase** also has the capability to catalyze the hydroxylation of various alkylated **aromatic** hydrocarbons. The low regiospecific hydroxylation was observed when toluene, o-xylene, ethyl benzene and n-propyl benzene were used as substrates.

=> s l3 and halogenated substrate
L8 0 L3 AND HALOGENATED SUBSTRATE

=> d his

(FILE 'HOME' ENTERED AT 11:43:23 ON 14 JAN 2003)

FILE 'MEDLINE, CAPLUS, BIOSIS, BIOTECHDS, SCISEARCH, EMBASE' ENTERED AT 11:44:23 ON 14 JAN 2003

L1 7515 S MONOOXYGENASE AND OXIDATION
L2 4386 DUP REM L1 (3129 DUPLICATES REMOVED)
L3 28 S L2 AND CYTOCHROME P-450CAM
L4 0 S L3 AND HOLOGENATED AROMATIC
L5 0 S L3 AND HALOGENATED AROMATIC
L6 0 S L3 AND HALO AROMATIC
L7 5 S L3 AND AROMATIC
L8 0 S L3 AND HALOGENATED SUBSTRATE

=> s l2 and halogenated aromatic substrate
2 FILES SEARCHED...
L9 0 L2 AND HALOGENATED AROMATIC SUBSTRATE

=> s l3 and dichlorobenzene
L10 1 L3 AND DICHLOROBENZENE

=> d l10 ibib ab

L10 ANSWER 1 OF 1 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2000:74430 CAPLUS

DOCUMENT NUMBER: 132:141329

TITLE: The **oxidation** of polychlorinated benzenes by genetically engineered cytochrome P450cam: potential applications in bioremediation

AUTHOR(S): Jones, Jonathan P.; O'Hare, Ellen J.; Wong, Luet-Lok

CORPORATE SOURCE: Dep. Chem., Inorganic Chem. Lab., University of Oxford, Oxford, OX1 3QR, UK

SOURCE: Chemical Communications (Cambridge) (2000), (3), 247-248

CODEN: CHCOFS; ISSN: 1359-7345

PUBLISHER: Royal Society of Chemistry

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Polychlorinated arom. compds. are persistent environmental pollutants.; we describe here. Redesign and engineering of the heme **monooxygenase**, **cytochrome P 450cam**, to oxidize these compds. efficiently to chlorinated phenols which are readily degraded by many microorganisms, thus provides a basis for novel bioremediation systems for these inert compds., are described.

REFERENCE COUNT: 13 THERE ARE 13 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

=> s l3 and (benzene or biphenyl)
L11 5 L3 AND (BENZENE OR BIPHENYL)

=> d 111 1-5 ibib ab

L11 ANSWER 1 OF 5 MEDLINE
ACCESSION NUMBER: 2002612268 MEDLINE
DOCUMENT NUMBER: 22241836 PubMed ID: 12114516
TITLE: Crystal structure of the F87W/Y96F/V247L mutant of
cytochrome P-450cam with
1,3,5-trichlorobenzene bound and further protein
engineering for the **oxidation** of
pentachlorobenzene and hexachlorobenzene.
AUTHOR: Chen Xuehui; Christopher Alexandra; Jones Jonathan P; Bell
Stephen G; Guo Qing; Xu Feng; Rao Zihe; Wong Luet-Lok
CORPORATE SOURCE: Laboratory of Structural Biology, Department of Biological
Science and Technology & Ministry of Education Laboratory
of Protein Science, Tsinghua University, Beijing 100084,
China.
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (2002 Oct 4) 277 (40)
37519-26.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
OTHER SOURCE: PDB-1J51
ENTRY MONTH: 200211
ENTRY DATE: Entered STN: 20021010
Last Updated on STN: 20030105
Entered Medline: 20021120

AB We reported previously that the F87W/Y96F/V247L mutant of
cytochrome P-450cam (CYP101) from *Pseudomonas*
putida catalyzed the rapid **oxidation** of lightly chlorinated
benzenes, but pentachlorobenzene **oxidation** was slow (Jones, J.
P., O'Hare, E. J., and Wong, L. L. (2001) *Eur. J. Biochem.* 268,
1460-1467). In the present work, we determined the crystal structure of
this mutant with bound 1,3,5-trichlorobenzene. The substrate was bound to
crystallographically independent CYP101 molecules in at least three
different orientations, which were distinguished by the angle between the
benzene ring and the porphyrin, and one orientation contained an
Fe-Cl interaction. In another orientation, the substrate was almost
parallel to the heme, with a C-H bond closest to the iron. The
enzyme/substrate contacts suggested that the L244A mutation should promote
the binding of pentachlorobenzene and hexachlorobenzene by creating space
to accommodate the extra chlorines. The F87W/Y96F/L244A/V247L mutant thus
designed was found to oxidize pentachlorobenzene at a rate of 82.5 nmol
(nmol CYP101)⁻¹ min⁻¹, 45 times faster than the F87W/Y96F/V247L parent
mutant. The rate of hexachlorobenzene **oxidation** was increased
200-fold, to 2.0 min⁻¹. Both substrates are oxidized to
pentachlorophenol, which is degraded by micro-organisms. In principle, the
F87W/Y96F/L244A/V247L mutant could have applications in the bioremediation
of polychlorinated benzenes.

L11 ANSWER 2 OF 5 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2002:220824 CAPLUS
DOCUMENT NUMBER: 136:259213
TITLE: Screening method for oxygenase enzymes using aromatic
substrates which are converted to spectrochemically
detected polymeric oxygenated compounds by a coupling
enzyme
INVENTOR(S): Arnold, Frances H.; Joo, Hyun
PATENT ASSIGNEE(S): California Institute of Technology, USA
SOURCE: PCT Int. Appl., 125 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent

LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
-----	----	-----	-----	-----
WO 2002022861	A1	20020321	WO 2000-US28768	20001013
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, EE, ES, FI, GB, GD, GE, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM				
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GN, GA, GN, GW, ML, MR, NE, SN, TD, TG				
AU 2000080285	A5	20020326	AU 2000-80285	20001013
PRIORITY APPLN. INFO.:			US 2000-661093	A1 20000913
			WO 2000-US28768	W 20001013
AB A method is provided for detecting the presence of an oxygenated compd. which is produced when a substrate is reacted with an oxygenase for the substrate. The method involves reacting a coupling enzyme with the oxygenated compd. to form a polymeric oxygenated compd. which is fluorescent or luminescent. Measurement of the fluorescence or luminescence of the polymeric oxygenated compd. provides indirect detection of the oxygenated compd. produced by reaction of the oxygenase with the substrate. The method is carried out in a whole cell environment wherein the cell is transformed to express both the oxygenase being screened and the coupling enzyme. The method can be used to measure the activity of monooxygenases and dioxygenases on arom. substrates. Thus, for example, the activity of cytochrome P 450cam in Escherichia coli is checked by measuring the conversion of naphthalene to a hydroxylated product (e.g., 1-naphthol, 2-naphthol) which emits a blue fluorescence when exogenously added horseradish peroxidase polymerizes the product. The method is amenable to large scale screening of enzyme mutants to isolate those with max. oxygenase activity. Thus, a screening strategy with high throughput fluorescence image anal. was implemented in order to identify bacterial clones expressing improved hydroxylating enzymes. Mutants of P 450cam with improved activity on naphthalene (or 3-phenylpropionate) and H2O2 are identified.				
REFERENCE COUNT:		2	THERE ARE 2 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT	

L11 ANSWER 3 OF 5 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 2000:911441 CAPLUS
DOCUMENT NUMBER: 134:68048
TITLE: Analogs of a cytochrome P450 of Pseudomonas putida with improved catalytic action aromatic halohydrocarbons for use in bioremediation of soil
INVENTOR(S): Wong, Luet Lok; Jones, Jonathan Peter
PATENT ASSIGNEE(S): Isis Innovation Limited, UK
SOURCE: PCT Int. Appl., 42 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
-----	----	-----	-----	-----
WO 2000078973	A1	20001228	WO 2000-GB2379	20000619
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR,				

HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT,
 LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU,
 SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN,
 YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
 RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY,
 DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ,
 CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG
 EP 1190067 A1 20020327 EP 2000-942200 20000619

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT,
 IE, SI, LT, LV, FI, RO

PRIORITY APPLN. INFO.: GB 1999-14373 A 19990618
 WO 2000-GB2379 W 20000619

AB Analogs of **cytochrome P 450cam** of
 Pseudomonas putida that have improved catalytic activity against heavily
 halogenated arom. hydrocarbons and that may be of use in the reclamation
 of soils contaminated with polychlorinated biphenyls. In particular,
 alterations in the substrate pocket that increase the vol. available for
 bulky polyhalogenated aroms. are described. Prepn. of a series of analogs
 of the gene camC cytochrome P 450 with increased activity towards
 polychlorinated biphenyls is demonstrated. A fusion protein of
 putidaredoxin and putidaredoxin reductase that can be used as a cofactor
 is also described.

REFERENCE COUNT: 7 THERE ARE 7 CITED REFERENCES AVAILABLE FOR THIS
 RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L11 ANSWER 4 OF 5 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2000:74430 CAPLUS

DOCUMENT NUMBER: 132:141329

TITLE: The **oxidation** of polychlorinated benzenes by
 genetically engineered cytochrome P450cam: potential
 applications in bioremediation

AUTHOR(S): Jones, Jonathan P.; O'Hare, Ellen J.; Wong, Luet-Lok

CORPORATE SOURCE: Dep. Chem., Inorganic Chem. Lab., University of
 Oxford, Oxford, OX1 3QR, UK

SOURCE: Chemical Communications (Cambridge) (2000), (3),
 247-248

CODEN: CHCOFS; ISSN: 1359-7345

PUBLISHER: Royal Society of Chemistry

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Polychlorinated arom. compds. are persistent environmental pollutants.; we
 describe here. Redesign and engineering of the heme **monooxygenase**
 , **cytochrome P 450cam**, to oxidize these
 compds. efficiently to chlorinated phenols which are readily degraded by
 many microorganisms, thus provides a basis for novel bioremediation
 systems for these inert compds., are described.

REFERENCE COUNT: 13 THERE ARE 13 CITED REFERENCES AVAILABLE FOR THIS
 RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L11 ANSWER 5 OF 5 SCISEARCH COPYRIGHT 2003 ISI (R)

ACCESSION NUMBER: 97:87565 SCISEARCH

THE GENUINE ARTICLE: WD134

TITLE: Gene organization and low regiospecificity in
 aromatic-ring hydroxylation of a **benzene**
monooxygenase of Pseudomonas aeruginosa J1104

AUTHOR: Kitayama A (Reprint); Suzuki E; Kawakami Y; Nagamune T

CORPORATE SOURCE: UNIV TOKYO, GRAD SCH ENGN, DEPT CHEM & BIOTECHNOL, BUNKYO
 KU, 7-3-1 HONGO, TOKYO 113, JAPAN (Reprint); INST RES &
 INNOVAT, DEPT BIOTECHNOL, KASHIWA, CHIBA 277, JAPAN

COUNTRY OF AUTHOR: JAPAN

SOURCE: JOURNAL OF FERMENTATION AND BIOENGINEERING, (20 JAN 1996)
 Vol. 82, No. 5, pp. 421-425.

Publisher: SOC FERMENTATION BIOENGINEERING, JAPAN, OSAKA

UNIV, FACULTY ENGINEERING, 2-1 YAMADAOKA, SUITA, OSAKA
565, JAPAN.
ISSN: 0922-338X.

DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE; AGRI
LANGUAGE: English
REFERENCE COUNT: 20

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB A novel **benzene monooxygenase** gene cluster has been
cloned from *Pseudomonas aeruginosa* J104 which was isolated from soil as a
benzene degrader. The nucleotide sequence of this gene cluster was
found to be highly homologous to those of other toluene
monooxygenase gene clusters. This multicomponent
monooxygenase also has the capability to catalyze the
hydroxylation of various alkylated aromatic hydrocarbons. The low
regiospecific hydroxylation was observed when toluene, o-xylene, ethyl
benzene and n-propyl **benzene** were used as substrates.

=> s 13 and (chlorine)
L12 1 L3 AND (CHLORINE)

=> d 112

L12 ANSWER 1 OF 1 MEDLINE
AN 93333178 MEDLINE
DN 93333178 PubMed ID: 7763853
TI Cosubstrate effects in reductive dehalogenation by *Pseudomonas putida* G786
expressing **cytochrome P-450CAM**.
AU Logan M S; Newman L M; Schanke C A; Wackett L P
CS Gray Freshwater Biological Institute, University of Minnesota, Navarre
55392.
NC GM41235 (NIGMS)
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TITLE: Cosubstrate effects in reductive dehalogenation by
Pseudomonas putida G786 expressing **cytochrome**
P-450CAM.
AUTHOR: Logan M S; Newman L M; Schanke C A; Wackett L P
CORPORATE SOURCE: Gray Freshwater Biological Institute, University of
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AB **Cytochrome P-450CAM** was shown to be the primary catalyst mediating reductive dehalogenation of polychlorinated ethanes by *Pseudomonas putida* G786. Under anaerobic conditions, the enzyme catalyzed reductive elimination reactions in vivo with the substrates hexachloroethane, pentachloroethane, and 1,1,1,2-tetrachloroethane; the products were tetrachloroethylene, trichloroethylene, and 1,1-dichloroethylene, respectively. In vivo reaction rates were determined. No reaction was observed with 1,1,2,2-tetrachloroethane or 1,1,1-trichloroethane. Purified **cytochrome P-450CAM** was used to measure dissociation constants of polychlorinated ethanes for the enzyme active site. Observed rates and dissociation constants were used to predict the course of a reaction with the three substrates simultaneously. Data obtained from experiments with *P. putida* G786 generally followed the simulated reaction curves. Oxygen suppressed the reductive dechlorination reactions and, in the case of 1,1,1,2-tetrachloroethane, 2,2,2-trichloroacetaldehyde was formed. Significant rates of reductive dechlorination were observed at 5% oxygen suggesting that these reactions could occur under partially aerobic conditions. These studies highlight the potential to use an aerobic bacterium, *P. putida* G786, under a range of oxygen tensions to reductively dehalogenate mixed wastes which are only degraded at very low rates by obligately anaerobic bacteria.

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(FILE 'HOME' ENTERED AT 11:43:23 ON 14 JAN 2003)

FILE 'MEDLINE, CAPLUS, BIOSIS, BIOTECHDS, SCISEARCH, EMBASE' ENTERED AT 11:44:23 ON 14 JAN 2003

L1	7515 S MONOOXYGENASE AND OXIDATION
L2	4386 DUP REM L1 (3129 DUPLICATES REMOVED)
L3	28 S L2 AND CYTOCHROME P-450CAM
L4	0 S L3 AND HOLOGENATED AROMATIC
L5	0 S L3 AND HALOGENATED AROMATIC
L6	0 S L3 AND HALO AROMATIC
L7	5 S L3 AND AROMATIC
L8	0 S L3 AND HALOGENATED SUBSTRATE
L9	0 S L2 AND HALOGENATED AROMATIC SUBSTRATE
L10	1 S L3 AND DICHLOBENZENE
L11	5 S L3 AND (BENZENE OR BIPHENYL)
L12	1 S L3 AND (CHLORINE)

=> log y

COST IN U.S. DOLLARS	SINCE FILE ENTRY	TOTAL SESSION
FULL ESTIMATED COST	93.26	93.68
DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)	SINCE FILE ENTRY	TOTAL SESSION
CA SUBSCRIBER PRICE	-3.91	-3.91

STN INTERNATIONAL LOGOFF AT 12:03:16 ON 14 JAN 2003

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L6: Entry 1 of 1

File: USPT

Oct 30, 2001

DOCUMENT-IDENTIFIER: US 6309883 B1

TITLE: Methods and compositions for cellular and metabolic engineering

Detailed Description Text (81):

Some examples of chemical targets for bioremediation include but are not limited to benzene, xylene, and toluene, camphor, naphthalene, halogenated hydrocarbons, polychlorinated biphenyls (PCBs), trichlorethylene, pesticides such as pentachlorophenyls (PCPs), and herbicides such as atrazine.

CLAIMS:

74. The method of claim 1, 2, or 3, wherein one or more member of the one or more sets of nucleic acids, the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid encodes one or more enzyme selected from the group consisting of: an acylase, a dioxygenase, a monooxygenase, a carotenoid synthetic enzyme, a hydrolytic enzyme, a catabolic enzyme, a dibenzothiophene catabolizing enzyme, a nitroreductase, a benzene degrading enzyme, a nitrobenzene degrading enzyme, a nitrotoluene degrading enzyme, a toxin degrading enzyme, an industrial chemical degrading enzyme, an herbicide degrading enzyme, a cellulose degrading enzyme, a pesticide degrading enzyme a pollutant degrading enzyme, a xylene degrading enzyme a toluene degrading enzyme, a camphor degrading enzyme, a naphthalene degrading enzyme, a halogenated hydrocarbon degrading enzyme, a biphenyl degrading enzyme, a polychlorinated biphenyl (PCB) degrading enzyme, a polycyclic aromatic hydrocarbon (PHA) degrading enzyme, a polyhydroxybutyrate (PHB) degrading enzyme, a trichlorethylene degrading enzyme, a pentachlorophenyl (PCP) degrading enzyme, a trichloroethylene degrading enzyme, a paranitrobenzyl, esterase, a sesquiterpene synthase, an expandase, a penicillin amidase, a penicillin G amidase, an enzyme which modifies 7-aminodeacetoxycephalosporanic acid (7-ADCA), an enzyme which modifies a semi-synthetically produced cephalosporin, and an enzyme which modifies penicillin V.

82. The method of claim 81, wherein the one or more toxin, industrial chemical, herbicide or pollutant comprises one or more of: benzene, xylene, toluene, camphor, naphthalene, a halogenated hydrocarbon, a polychlorinated biphenyl (PCB), a polycyclic aromatic hydrocarbon (PHA), a trichlorethylene, a pentachlorophenyl (PCP) or trichloroethylene.

103. The method of claim 102, the one or more toxin, industrial chemical, herbicide or pollutant comprising one or more of: benzene, xylene, toluene, camphor, naphthalene, a halogenated hydrocarbon, a polychlorinated biphenyl (PCB), a polycyclic aromatic hydrocarbon (PHA), a trichlorethylene, a pentachlorophenyl (PCP) or trichloroethylene.

126. The method of claim 1 or 3, wherein the screening comprises monitoring one or more enzymatic activities of one or more enzymes selected from the group consisting of: an acylase, a dioxygenase, a monooxygenase, a carotenoid synthetic enzyme, a hydrolytic enzyme, a catabolic enzyme, a nitroreductase, a benzene degrading enzyme, a nitrobenzene degrading enzyme, a nitrotoluene degrading enzyme, a toxin degrading enzyme, an industrial chemical degrading enzyme, an herbicide degrading enzyme, a cellulose degrading enzyme, a pesticide degrading enzyme a pollutant degrading enzyme, a xylene degrading enzyme, a toluene degrading enzyme, a camphor degrading enzyme, a naphthalene degrading enzyme, a halogenated hydrocarbon degrading enzyme, a polychlorinated biphenyl (PCB) degrading enzyme, a polycyclic aromatic hydrocarbon (PHA) degrading enzyme, a polyhydroxybutyrate (PHP) degrading enzyme, a trichlorethylene degrading enzyme, a pentachlorophenyl (PCP) degrading enzyme, a

trichloroethylene degrading enzyme, a paranitrobenzyl, esterase, a sesquiterpene synthase, an expandase, a penicillin amidase, a penicillin G amidase, an enzyme which modifies 7-aminodeacetoxycephalosporanic acid (7-ADCA), an enzyme which modifies a semi-synthetically produced cephalosporin, and an enzyme which modifies penicillin V.

128. The method of claim 127 wherein the one or more toxin, industrial chemical, herbicide or pollutant comprises one or more of: benzene, xylene, toluene, camphor, naphthalene, a halogenated hydrocarbon, a polychlorinated biphenyl (PCB), a polycyclic aromatic hydrocarbon (PHA), a trichlorethylene, a pentachlorophenyl (PCP) or trichloroethylene.

WEST**End of Result Set**

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L6: Entry 1 of 1

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TITLE: Methods and compositions for cellular and metabolic engineering

DATE-ISSUED: October 30, 2001

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
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US-CL-CURRENT: 435/440; 435/6, 536/23.1, 536/24.3

CLAIMS:

What is claimed is:

1. A method of recombining one or more nucleic acids, the method comprising:

introducing one or more sets of nucleic acids into a plurality of cells, thereby providing a plurality of modified cells, each of the plurality of modified cells comprising at least one member of the one or more sets of nucleic acids;

transferring at least a first member of the one or more sets of nucleic acids from a first of the plurality of modified cells into at least a second of the plurality of modified cells;

permitting recombination to occur between the at least one member of the one or more sets of nucleic acids and a nucleic acid present in the second of the plurality of modified cells to produce a recombinant nucleic acid;

introducing the recombinant nucleic acid into a third cell and permitting recombination between the recombinant nucleic acid and a third member present in a third cell of the plurality of modified cells, or between the recombinant nucleic acid and the first member or the second member, thereby producing a further recombined nucleic acid; and,

screening the further recombined nucleic acid for one or more properties or one or more encoded activities, thereby providing a selected recombinant nucleic acid.

2. The method of claim 1, comprising further recombining the selected recombinant nucleic acid with one or more additional nucleic acids and selecting the resulting further recombined nucleic acid to produce a further recombined selected nucleic acid.

3. A method of recombining one or more nucleic acids, the method comprising:

introducing one or more sets of nucleic acids into a plurality of cells, thereby providing a plurality of modified cells, each of the plurality of modified cells comprising at least one member of the one or more sets of nucleic acids;

transferring at least a first member of the one or more sets of nucleic acids from a first of the plurality of modified cells into at least a second of the plurality of modified cells; permitting recombination to occur between the first member and a second member present in the second of the plurality of modified cells, thereby producing a recombinant nucleic acid;

screening the recombinant nucleic acid for one or more properties or one or more encoded activities; and,

further recombining the selected recombinant nucleic acid with one or more additional nucleic acid, or with the first or second nucleic acid, thereby producing a further recombined selected nucleic acid.

4. The method of claim 2 or 3, comprising screening the further recombined selected nucleic acid for one or more encoded activities, thereby providing a multiply recombined multiply selected nucleic acid.

5. The method of claim 2 or 3, wherein the further recombining comprises in vitro recombination.

6. The method of claim 5, wherein the further recombining comprises recursive in vitro recombination.

7. The method of claim 2 or 3, wherein the further recombining comprises in vivo recombination.

8. The method of claim 7, wherein the further recombining step comprises recursive in vivo recombination.

9. The method of claim 1 or 3, wherein the one or more sets of nucleic acids comprise one or more nucleic acid produced by in vitro sequence recombination.

10. The method of claim 1 or 3, wherein the one or more sets of nucleic acids comprise one or more nucleic acid produced by recursive in vitro recombination.

11. The method of claim 1 or 3, wherein the one or more sets of nucleic acids comprise one or more nucleic acid produced by in vivo recombination.

12. The method of claim 1 or 3, wherein the one or more sets of nucleic acids comprise one or more nucleic acid produced by recursive in vivo sequence recombination.

13. The method of claim 1 or 3, wherein the one or more sets of nucleic acids comprise one or more nucleic acid produced by mutation.

14. The method of claim 13, wherein the one or more sets of nucleic acids are produced by error prone PCR.

15. The method of claim 1 or 3, wherein the step of introducing the one or more sets of nucleic acids into the plurality of cells comprises packaging members of one or more of the one or more sets into phage vectors and transducing the resulting phage library into a plurality of cells, thereby producing the plurality of modified cells.

16. The method of claim 1 or 3, wherein the step of introducing the one or more sets of nucleic acids into the plurality of cells comprises packaging members of one or more of the one or more sets into viral vectors and transducing the

resulting viral library into a plurality of cells, thereby producing the plurality of modified cells.

17. The method of claim 1 or 3, wherein the step of introducing the one or more sets of nucleic acids into the plurality of cells comprises electroporating members of one or more of the one or more sets into a plurality of cells, thereby producing the plurality of modified cells.

18. The method of claim 1 or 3, wherein the step of introducing the one or more sets of nucleic acids into the plurality of cells comprises electronic pulse introduction of members of one or more of the one or more sets into a plurality of cells, thereby producing the plurality of modified cells.

19. The method of claim 1 or 3, wherein the step of introducing the one or more sets of nucleic acids into the plurality of cells comprises biolistically introducing members of one or more of the one or more sets into a plurality of cells, thereby producing the plurality of modified cells.

20. The method of claim 1 or 3, wherein the step of introducing the one or more sets of nucleic acids into the plurality of cells comprises transferring members of one or more of the one or more sets into a plurality of cells via conjugative transfer, thereby producing the plurality of modified cells.

21. The method of claim 1 or 3, wherein the step of introducing the one or more sets of nucleic acids into the plurality of cells comprises transferring one or more of the one or more sets into the plurality of cells by fusing one or more cells comprising one or more members of the one or more sets with a plurality of cells, thereby producing the plurality of modified cells.

22. The method of claim 1 or 3, wherein the step of introducing the one or more sets of nucleic acids into the plurality of cells comprises transferring one or more members of the one or more sets of nucleic acids into a plurality of cells by fusing one or more library cells comprising members of the one or more sets with the one or more of the plurality of cells, wherein the fusing is induced by incubation of the library cells or the plurality of cells, or both, with a viral protein, or a chemical agent.

23. The method of claim 22, wherein the viral protein comprises one or more of: an influenza protein, an influenza viral hemagglutinin protein, HSV-1 g B, or HSV-1 g D.

24. The method of claim 22, wherein the chemical agent is PEG.

25. The method of claim 1 or 3, wherein the step of transferring at least a first member of the one or more sets of nucleic acids from a first of the plurality of modified cells into at least a second of the plurality of modified cells comprises packaging at least one member of one or more of the one or more sets into at least one phage vector and transducing the resulting at least one phage vector into the second modified cell.

26. The method of claim 1 or 3, wherein the step of transferring at least a first member of the one or more sets of nucleic acids from a first of the plurality of modified cells into at least a second of the plurality of modified cells comprises packaging at least one member of one or more of the one or more sets into at least one viral vector and transducing the resulting at least one viral vector into the second modified cell.

27. The method of claim 1 or 3, wherein the step of transferring at least a first member of the one or more sets of nucleic acids from a first of the plurality of modified cells into at least a second of the plurality of modified cells comprises electroporating at least one member of one or more of the one or more sets into the second modified cell.

28. The method of claim 1 or 3, wherein the step of transferring at least a first member of the one or more sets of nucleic acids from a first of the plurality of modified cells into at least a second of the plurality of modified cells comprises electronic pulse transfer of at least one member of one or more of the one or more sets into the second modified cell.

29. The method of claim 1 or 3, wherein the step of transferring at least a first member of the one or more sets of nucleic acids from a first of the plurality of modified cells into at least a second of the plurality of modified cells comprises biolistically transferring at least one member of one or more of the one or more sets into the second modified cell.

30. The method of claim 1 or 3, wherein the step of transferring at least a first member of the one or more sets of nucleic acids from a first of the plurality of modified cells into at least a second of the plurality of modified cells is performed via conjugative transfer of the first member from the first modified cell into the second modified cell.

31. The method of claim 1 or 3, wherein the step of transferring at least a first member of the one or more sets of nucleic acids from a first of the plurality of modified cells into at least a second of the plurality of modified cells is performed by fusing the first and second cell.

32. The method of claim 1 or 3, wherein the step of transferring at least a first member of the one or more sets of nucleic acids from a first of the plurality of modified cells into at least a second of the plurality of modified cells is performed by fusing the first and second cell, wherein the fusing is induced by incubation of the first and second cells with a viral protein, or a chemical agent.

33. The method of claim 32, wherein the viral protein comprises one or more of: an influenza protein, an influenza viral hemagglutinin protein, HSV-1 g B, or HSV-1 g D.

34. The method of claim 32, wherein the chemical agent is PEG.

35. The method of claim 1 or 3, wherein the step of introducing the recombinant nucleic acid into the third cell comprises packaging the recombinant nucleic acid into at least one phage vector and transducing the resulting at least one phage vector into the third cell.

36. The method of claim 1 or 3, wherein the step of introducing the recombinant nucleic acid into the third cell comprises packaging the recombinant nucleic acid into at least one viral vector and transducing the resulting at least one viral vector into the third cell.

37. The method of claim 1 or 3, wherein the step of introducing the recombinant nucleic acids into the third cell comprises electroporating the recombinant nucleic acid into the third cell.

38. The method of claim 1 or 3, wherein the step of introducing the recombinant nucleic acid into the third cell comprises pulse introducing the recombinant nucleic acid into the third cell.

39. The method of claim 1 or 3, wherein the step of introducing the recombinant nucleic acid into the third cell comprises biolistically introducing the recombinant nucleic acid into the third cell.

40. The method of claim 1 or 3, wherein the step of introducing the recombinant nucleic acid into the third cell is performed via conjugative transfer of the recombinant nucleic acid into the third cell.

41. The method of claim 1 or 3, wherein the step of introducing the recombinant nucleic acid into the third cell comprises fusing the second and third cells.

42. The method of claim 1 or 3, wherein the step of introducing the recombinant nucleic acid into the third cell comprises fusing the second and third cells, wherein the fusing is induced by incubation of the second and third cells with a viral protein, or a chemical agent.

43. The method of claim 42, wherein the viral protein comprises one or more of: an influenza protein, an influenza viral hemagglutinin protein, HSV-1 g B, or HSV-1 g D.

44. The method of claim 42, wherein the chemical agent is PEG.

45. The method of claim 1 or 3, wherein the plurality of modified cells comprise one or more mutator cells.

46. The method of claim 45, wherein the mutator cells are selected from the group consisting of: Mut L cells, Mut S cells, Mut D cells, Mut T cells, Mut H cells, and Human Ataxia Telangiectia cells.

47. The method of claim 1 or 3, wherein a plurality of members of the one or more sets of nucleic acids are at least about 50% identical.

48. The method of claim 1 or 3, wherein the members of the one or more sets of nucleic acids are at least about 70% identical.

49. The method of claim 1 or 3, wherein the members of the one or more sets of nucleic acids are at least about 80% identical.

50. The method of claim 1 or 3, wherein the members of the one or more sets of nucleic acids are at least about 90% identical.

51. The method of claim 1 or 3, wherein the members of the one or more sets of nucleic acids differ from each other in about 5 to about 20 positions.

52. The method of claim 1 or 3, wherein at least one of the one or more sets of nucleic acids have less than 10 members.

53. The method of claim 1 or 3, wherein at least one of the one or more sets of nucleic acids have more than 10.sup.5 members.

54. The method of claim 1 or 3, wherein at least one of the one or more sets of nucleic acids have more than 10.sup.7 members.

55. The method of claim 1 or 3, wherein at least one of the one or more sets of nucleic acids have more than 10.sup.9 members.

56. The method of claim 1 or 3, wherein at least one member of the one or more sets of nucleic acids is a full-length gene.

57. The method of claim 1 or 3, wherein at least one member of the one or more sets of nucleic acids is cloned into a vector which supplies one or more of: a promoter, a polyadenylation sequence, or a regulatory sequence.

58. The method of claim 1 or 3, wherein the members of the one or more sets of nucleic acids are allelic or species variants.

59. The method of claim 1 or 3, wherein at least one member of the plurality of modified cells is selected or derived from one or more of: a bacterial cell, gram-negative cell, gram-positive cell, a Streptomyces cell, an Actinomycetes cell, a Corynebacteria cell, a Penicillium cell, a Bacillus cell, an

Escherichia coli cell, a Pseudomonas cell, a Salmonella cell, an Erwinia cell, a eukaryotic cell, a mammalian cell, a mouse cell, a hamster cell, a primate cell, a human cell, an established cell line cell, a primary cell culture cell, a stem cell, an embryonic stem cell, a zygotes cell, a fibroblast cell, a lymphocyte cell, a Chinese hamster ovary (CHO) cell, a mouse fibroblast cell, an NIH3T3 cell, a kidney cell, a liver cell, a muscle cell, a skin cell, a plant cell, a maize cell, a rice cell, a wheat cell, a cotton cell, a soybean cell, a sugarcane cell, a tobacco cell, an arabidopsis cell; a fish cell, an algal cell, a fungal cell, a Penicillium cell, a Fusarium cell, an Aspergillus cell, a Podospora cell, a Neurospora cell, an insect cell, a yeast cell, a Picchia cell, a Saccharomyces cell, or a nitrogen-fixation symbiotic cell.

60. The method of claim 1 or 3, wherein at least one member of the plurality of modified cells is selected or derived from a tissue or organism selected from the group consisting of: a plant, a bacteria, a fungus, an algae, an intact animal tissue, a tissue culture, and an animal embryo.

61. The method of claim 1 or 3, wherein at least one member of the plurality of modified cells is selected or derived from one or more of: E. coli, lactobacilli, Streptomyces, Actinomyces or filamentous fungi.

62. The method of claim 1 or 3, wherein at least one member of the plurality of modified cells is selected for one or more of: pathogenicity, substrate range, environmental hardiness, presence of one or more key intermediates, ease of genetic manipulation, or likelihood of promiscuous transfer of genetic information to other organisms.

63. The method of claim 1 or 3, wherein at least one member of the plurality of modified cells is selected or derived from one or more cell which comprises a biphenyl catabolizing pathway.

64. The method of claim 1, 2, or 3, wherein one or more member of the one or more sets of nucleic acids, the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid comprises one or more of: a plasmid, a cosmid, a chromosome, an episome, a YAC, a phage, a filamentous phage, a phage P1 clone, or a viral vector.

65. The method of claim 1, 2, or 3, wherein one or more member of the one or more sets of nucleic acids, the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid comprises cleaved genomic DNA.

66. The method of claim 1, 2, or 3, wherein one or more member of the one or more sets of nucleic acids, the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid comprises amplified genomic DNA.

67. The method of claim 1, 2, or 3, wherein one or more member of the one or more sets of nucleic acids, the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid comprises one or more metabolic pathway nucleic acids which encode at least one metabolic pathway.

68. The method of claim 1, 2, or 3, wherein one or more member of the one or more sets of nucleic acids, the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid are provided in or selected from a library of nucleic acids selected from the group consisting of: a plasmid library, a cosmid library, a phage library, a chromosome library, a filamentous phage library, and a viral library.

69. The method of claim 1, 2, or 3, wherein one or more member of the one or more sets of nucleic acids, the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid are provided in or selected from a library of nucleic acids comprising variants of a single gene.

70. The method of claim 1, 2, or 3, wherein one or more member of the one or more sets of nucleic acids, the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid are provided in or selected from a library of nucleic acids comprising variants of more than one gene.

71. The method of claim 1, 2, or 3, wherein one or more member of the one or more sets of nucleic acids, the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid are provided in or selected from a library of nucleic acids comprising one or more genes in a biochemical pathway.

72. The method of claim 1, 2, or 3, wherein one or more member of the one or more sets of nucleic acids, the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid are provided in or selected from a library of genes isolated from one or more of: a bacteria, an *Alcaligenes*, a *Zoogloea*, a *Rhizobium*, a *Bacillus*, a *Azobacter*, or a eukaryote.

73. The method of claim 1, 2, or 3, wherein one or more member of the one or more sets of nucleic acids, the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid comprises a nucleic acid which encodes a regulatory gene.

74. The method of claim 1, 2, or 3, wherein one or more member of the one or more sets of nucleic acids, the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid encodes one or more enzyme selected from the group consisting of: an acylase, a dioxygenase, a monooxygenase, a carotenoid synthetic enzyme, a hydrolytic enzyme, a catabolic enzyme, a dibenzothiophene catabolizing enzyme, a nitroreductase, a benzene degrading enzyme, a nitrobenzene degrading enzyme, a nitrotoluene degrading enzyme, a toxin degrading enzyme, an industrial chemical degrading enzyme, an herbicide degrading enzyme, a cellulose degrading enzyme, a pesticide degrading enzyme, a pollutant degrading enzyme, a xylene degrading enzyme, a toluene degrading enzyme, a camphor degrading enzyme, a naphthalene degrading enzyme, a halogenated hydrocarbon degrading enzyme, a biphenyl degrading enzyme, a polychlorinated biphenyl (PCB) degrading enzyme, a polycyclic aromatic hydrocarbon (PHA) degrading enzyme, a polyhydroxybutyrate (PHB) degrading enzyme, a trichlorethylene degrading enzyme, a pentachlorophenyl (PCP) degrading enzyme, a trichloroethylene degrading enzyme, a paranitrobenzyl, esterase, a sesquiterpene synthase, an expandase, a penicillin amidase, a penicillin G amidase, an enzyme which modifies 7-aminodeacetoxycephalosporanic acid (7-ADCA), an enzyme which modifies a semi-synthetically produced cephalosporin, and an enzyme which modifies penicillin V.

75. The method of claim 74, wherein the enzyme is a polyhydroxybutyrate (PHB) degrading enzyme, wherein the one or more sets of nucleic acids are derived from one or more of: an *Alcaligenes* bacteria, a *Zoogloea* bacteria, a *Rhizobium* bacteria, a *Bacillus* bacteria, or an *Azobacter* bacteria.

76. The method of claim 74, wherein the enzyme is a biphenyl degrading enzyme and wherein the enzyme is expressed in at least one host cell which comprises a biphenyl catabolizing pathway.

77. The method of claim 74, wherein the enzyme is a cellulose degrading enzyme and wherein the one or more sets of nucleic acids are derived from one or more *Agrobacterium tumefaciens*.

78. The method of claim 74, wherein the enzyme is a carotenoid synthetic enzyme and wherein the one or more sets of nucleic acids are derived from one or more of: *Myxococcus xanthus*, *Rhodobacter sphaeroides*, *Thermus thermophilus*, *Erwinia uredovora*, *Haematococcus pluvialis*, *E. coli*, *E. herbicola*, or *R. capsulatus*.

79. The method of claim 1, 2, or 3, wherein one or more member of the one or more sets of nucleic acids, the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid encodes one or more enzyme which is resistant to inactivation by one or more epoxide.

80. The method of claim 1, 2, or 3, wherein the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid provides one or more organism, when expressed in the organism, with a new or improved ability to convert a pollutant into a nutrient source.

81. The method of claim 1, 2, or 3, wherein the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid provides one or more organism, when expressed in the organism, with a new or improved ability to degrade one or more toxin, industrial chemical, herbicide, pesticide or pollutant.

82. The method of claim 81, wherein the one or more toxin, industrial chemical, herbicide or pollutant comprises one or more of: benzene, xylene, toluene, camphor, naphthalene, a halogenated hydrocarbon, a polychlorinated biphenyl (PCB), a polycyclic aromatic hydrocarbon (PHA), a trichlorethylene, a pentachlorophenyl (PCP) or trichloroethylene.

83. The method of claim 1, 2, or 3, wherein the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid encode an enzyme with an improved catalytic activity, a new catalytic activity, altered substrate recognition, thermostability, stability in a non-aqueous solvent, or an altered expression level.

84. The method of claim 1, 2, or 3, wherein the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid provides one or more organism, when expressed in the organism, with a new or improved resistance to the presence of one or more heavy metal.

85. The method of claim 1, 2, or 3, wherein the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid provides one or more organism, when expressed in the organism one or more property selected from the group consisting of: modified growth rate, ability to secrete a desired compound, an ability to tolerate an increased temperature, and an ability to tolerate one or more environmental stress.

86. The method of claim 1, 2, or 3, wherein the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid provides one or more organism, when expressed in the organism, with a new or improved ability to reduce an organo-nitro compound or to permit the organism to survive in the presence of an organo-nitro compound.

87. The method of claim 1, 2, or 3, wherein the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid provides one or more organism, when expressed in the organism, with new or improved utilization of a nutrient source.

88. The method of claim 87, wherein the nutrient source is selected from the group consisting of: lactose, whey, galactose, mannitol, xylan, cellobiose, cellulose and sucrose.

89. The method of claim 87, wherein the improved utilization of a nutrient source provides for production of compounds selected from the group consisting of: ethanol, tryptophan, a rhamnolipid surfactant, xanthan gum, polysaccharide xanthan gum and polyhydroxylalkanoate.

90. The method of claim 1, 2, or 3, wherein the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid provides one or more organism, when expressed in the organism, new or improved production of one or more product selected from the group consisting of: ethanol, tryptophan, a rhamnolipid surfactant, xanthan gum, polysaccharide xanthan gum, polyhydroxylalkanoate, phenylalanine, and 2-keto-L-gluconic acid.

91. The method of claim 1, 2, or 3, wherein the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid provides one or more organism, when expressed in the organism, with a new or improved ability to produce one or more metabolic intermediate.

92. The method of claim 91, wherein the metabolic intermediate is selected from the group consisting of: an antibiotic, a vitamin, an amino acid, phenylalanine, an aromatic amino acid, ethanol, butanol, polysaccharide xanthan gum, xanthan gum, bacterial cellulose, a peptide, and a lipid.

93. The method of claim 1, 2, or 3, wherein the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid encodes an enzyme which produces one or more compound selected from the group consisting of: a polyketide, a dye, a vitamin, an antibiotic, a carotenoid, a terpenoid, and an isoprenoid.

94. The method of claim 93, wherein the dye is indigo.

95. The method of claim 93, wherein the vitamin is vitamin C.

96. The method of claim 93, wherein the antibiotic is selected from the group consisting of: a peptide, a peptidolactone, a thiopeptide, a beta-lactam, a glycopeptide, a lantibiotic, a microcin, a polyketide-derived antibiotic, an anthracyclin, a tetracyclin, a macrolide, an avermectin, a polyether, an ansamycins, chloramphenicol, an aminoglycoside, an aminocyclitol, a polyoxin, an agrocin, mederrhodin, dihydrogranatirhodin, 6-deoxyerythromycin A, isovalerylspiramycin, a hybrid macrolide and an isoprenoid.

97. The method of claim 93, wherein the polyketide is an antibiotic.

98. The method of claim 93, wherein the polyketide is selected from the group consisting of: tetracycline, erythromycin, an anti-cancer agent, daunomycin, an immunosuppressant, FK506, rapamycin, monesin and avermectin.

99. The method of claim 93, wherein the isoprenoid is selected from the group consisting of: an antibacterial isoprenoid and an antifungal isoprenoid.

100. The method of claim 93, wherein the carotinoid is selected from the group consisting of: a ketocarotenoid, a myxobacton, a spheroidene, a spheroidenone, a lutein, an astaxanthin, a violaxanthin, a 4-ketorulene, a myxoxanthrophyll, an echinenone, a lycopene, a zeaxanthin, a monoglucoside, a diglucoside, an alpha carotene, a beta carotene, a gamma carotene, a delta carotene, a cryptoxanthin monoglucoside and a neoxanthin.

101. The method of claim 1 or 3, further comprising propagating the first, second, or third cell, in culture.

102. The method of claim 1 or 3, wherein the screening comprises monitoring bioremediation or biodegradation of one or more toxin, industrial chemical, herbicide, pesticide or pollutant.

103. The method of claim 102, the one or more toxin, industrial chemical, herbicide or pollutant comprising one or more of: benzene, xylene, toluene, camphor, naphthalene, a halogenated hydrocarbon, a polychlorinated biphenyl (PCB), a polycyclic aromatic hydrocarbon (PHA), a trichlorethylene, a pentachlorophenyl (PCP) or trichloroethylene.

104. The method of claim 1 or 3, wherein the screening step is performed in the same cell type as the recombinant cell is produced in.

105. The method of claim 1 or 3, wherein the screening step is performed in a different cell type than the recombinant cell is produced in.

106. The method of claim 1 or 3, wherein the screening comprises monitoring one or more reporter gene selected from the group consisting of: luciferase, green fluorescence protein, and .beta.-galactosidase.

107. The method of claim 1 or 3, wherein the screening comprises monitoring one or more of: fluorescence, bioluminescence, colony size, cell growth rate, a chromogenic substrate, or halo formation.

108. The method of claim 1 or 3, wherein the screening comprises performing an ELISA assay.

109. The method of claim 1 or 3, wherein the screening comprises performing a cell-cell activity assay.

110. The method of claim 2 or 3, wherein the screening comprises monitoring differential expression of a protein or nucleic acid expressed in a screened cell comprising the recombinant nucleic acid, the further recombined nucleic acid, or the further recombined selected nucleic acid.

111. The method of claim 1 or 3, wherein the screening comprises performing FACS.

112. The method of claim 1 or 3, wherein the screening comprises performing two-color FACS.

113. The method of claim 1 or 3, wherein the screening comprises monitoring gel microdroplets.

114. The method of claim 1 or 3, wherein the screening comprises detecting one or more molecule by mass spectrometry.

115. The method of claim 1, 2, or 3, wherein a selected cell comprising the recombinant nucleic acid, the further recombined nucleic acid, the further recombined selected nucleic acid or the multiply recombined multiply selected nucleic acid is selected in a chemostat.

116. The method of claim 1 or 3, wherein the screening comprises selecting for one or more of: an improved catalytic activity, a new catalytic activity, altered substrate recognition, thermostability, stability in a non-aqueous solvent, or an altered expression level.

117. The method of claim 1 or 3, wherein the screening comprises selecting one or more organism comprising the recombinant nucleic acid for one or more property selected from the group consisting of: a modified growth rate, an ability to secrete a desired compound, an ability to tolerate an increased temperature, and an ability to tolerate one or more environmental stresses.

118. The method of claim 1 or 3, wherein the screening comprises monitoring the presence or absence of one or more secondary metabolite selected from the group consisting of: a polyketide, a dye, a vitamin, an antibiotic, a carotenoid, a terpenoid, and an isoprenoid.

119. The method of claim 118, wherein the dye is indigo.

120. The method of claim 118, wherein the vitamin is vitamin C.

121. The method of claim 118, wherein the antibiotic is selected from the group consisting of: a peptide, a peptidolactone, a thiopeptide, a beta-lactam, a glycopeptide, a lantibiotic, a microcin, a polyketide-derived antibiotic, an anthracyclin, a tetracyclin, a macrolide, an avermectin, a polyether, an ansamycins, chloramphenicol, an aminoglycoside, an aminocyclitol, a polyoxin, an agrocin, mederrhodin, dihydrogranatirhodin, 6-deoxyerythromycin A, isovalerylspiramycin, a hybrid macrolide and an isoprenoid.

122. The method of claim 118, wherein the polyketide is an antibiotic.

123. The method of claim 118, wherein the polyketide is selected from the group consisting of: tetracycline, erythromycin, an anti-cancer agent, daunomycin, an immunosuppressant, FK506, rapamycin, monesin and avermectin.

124. The method of claim 118, wherein the isoprenoid is selected from the group consisting of: an antibacterial isoprenoid and an antifungal isoprenoid.

125. The method of claim 118, wherein the carotenoid is selected from the group consisting of: a ketocarotenoid, a myxobacton, a spheroidene, a spheroidenone, a lutein, an astaxanthin, a violaxanthin, a 4-ketorulene, a myxoxanthrophyll, an echinenone, a lycopene, a zeaxanthin, a monoglucoside, a diglucoside, an alpha carotene, a beta carotene, a gamma carotene, a delta carotene, a cryptoxanthin monoglucoside and a neoxanthin.

126. The method of claim 1 or 3, wherein the screening comprises monitoring one or more enzymatic activities of one or more enzymes selected from the group consisting of: an acylase, a dioxygenase, a monooxygenase, a carotenoid synthetic enzyme, a hydrolytic enzyme, a catabolic enzyme, a nitroreductase, a benzene degrading enzyme, a nitrobenzene degrading enzyme, a nitrotoluene degrading enzyme, a toxin degrading enzyme, an industrial chemical degrading enzyme, an herbicide degrading enzyme, a cellulose degrading enzyme, a pesticide degrading enzyme, a pollutant degrading enzyme, a xylene degrading enzyme, a toluene degrading enzyme, a camphor degrading enzyme, a naphthalene degrading enzyme, a halogenated hydrocarbon degrading enzyme, a polychlorinated biphenyl (PCB) degrading enzyme, a polycyclic aromatic hydrocarbon (PHA) degrading enzyme, a polyhydroxybutyrate (PHP) degrading enzyme, a trichlorethylene degrading enzyme, a pentachlorophenyl (PCP) degrading enzyme, a trichloroethylene degrading enzyme, a paranitrobenzyl, esterase, a sesquiterpene synthase, an expandase, a penicillin amidase, a penicillin G amidase, an enzyme which modifies 7-aminodeacetoxycephalosporanic acid (7-ADCA), an enzyme which modifies a semi-synthetically produced cephalosporin, and an enzyme which modifies penicillin V.

127. The method of claim 1 or 3, wherein the screening comprises monitoring degradation of one or more of: a toxin, an industrial chemical, an herbicide, a pesticide a pollutant, PHB, or cellulose.

128. The method of claim 127 wherein the one or more toxin, industrial chemical, herbicide or pollutant comprises one or more of: benzene, xylene, toluene, camphor, naphthalene, a halogenated hydrocarbon, a polychlorinated biphenyl (PCB), a polycyclic aromatic hydrocarbon (PHA), a trichlorethylene, a pentachlorophenyl (PCP) or trichloroethylene.

129. The method of claim 1 or 3, wherein the screening comprises monitoring synthesis of one or more carotenoid.

130. The method of claim 1 or 3, wherein the screening comprises monitoring resistance of an enzyme to an epoxide.

131. The method of claim 1 or 3, wherein the screening comprises monitoring resistance of a cell modified with the recombinant nucleic acid to a heavy metal.

132. The method of claim 1 or 3, wherein the screening comprises selecting an organism which expresses the recombinant nucleic acid for an ability to survive in the presence of an organo-nitro compound.

133. The method of claim 1 or 3, wherein the screening comprises selecting an organism for an ability to metabolize lactose, whey, galactose, mannitol, xylan, cellobiose, cellulose or sucrose.

134. The method of claim 1 or 3, wherein the screening comprises selecting, an organism for an ability to produce ethanol, tryptophan, a rhamnolipid surfactant, xanthan gum, polysaccharide xanthan gum, polyhydroxylalkanoate, phenylalanine, or 2-keto-L-gluconic acid.

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L5: Entry 1 of 7

File: USPT

Apr 23, 2002

US-PAT-NO: 6376254

DOCUMENT-IDENTIFIER: US 6376254 B1

TITLE: Biomimetic reagent system and its use

DATE-ISSUED: April 23, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Bather; Wolfgang	Lubeck			DE
Duchstein; Hans-Jurgen	Pinneberg			DE
Hoffmann; Susanne	Buchholz			DE

ASSIGNEE-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY	TYPE CODE
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APPL-NO: 09/ 394969 [PALM]

DATE FILED: September 10, 1999

FOREIGN-APPL-PRIORITY-DATA:

COUNTRY	APPL-NO	APPL-DATE
DE	199 12 380	March 19, 1999

INT-CL: [07] G01 N 21/78

US-CL-ISSUED: 436/140; 436/167, 422/86, 422/88

US-CL-CURRENT: 436/140; 422/86, 422/88, 436/167

FIELD-OF-SEARCH: 436/140, 436/164, 436/167, 436/169, 436/181, 422/55, 422/86, 422/87, 422/88, 422/91

PRIOR-ART-DISCLOSED:

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ART-UNIT: 1743

PRIMARY-EXAMINER: Snay; Jeffrey

ABSTRACT:

A biomimetic reagent system is provided containing an oxygen donor and a catalyst based on porphyrin, which are applied to a carrier. A device that contains the system is also provided for determining components of gas or vapor samples, especially aromatics, such as benzene. A process for hydroxylating aromatics, such as benzene, using the biomimetic reagent system is also provided.

30 Claims, 1 Drawing figures

WEST**End of Result Set**

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L6: Entry 1 of 1

File: USPT

Oct 30, 2001

US-PAT-NO: 6309883

DOCUMENT-IDENTIFIER: US 6309883 B1

TITLE: Methods and compositions for cellular and metabolic engineering

DATE-ISSUED: October 30, 2001

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Minshull; Jeremy	San Francisco	CA		
Stemmer; Willem P. C.	Los Gatos	CA		

ASSIGNEE-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY	TYPE CODE
Maxygen, Inc.	Redwood City	CA			02

APPL-NO: 09/ 490642 [PALM]

DATE FILED: January 24, 2000

PARENT-CASE:

This application is a CON of Ser. No. 09/189,103 filed Nov. 9, 1998, which is a CON of Ser. No. 08/650,400 filed May 20, 1996, now U.S. Pat. No. 5,837,458; which is a CIP of Ser. No. 08/198,431 filed Feb. 17, 1994, now U.S. Pat. No. 5,605,793; and a CIP of Ser. No. 08/621,859 filed Mar. 25, 1996, now U.S. Pat. No. 6,117,679; and a CIP of Ser. No. 08/621,430 filed Mar. 25, 1996, now abandoned; and a CIP of Ser. No. 08/537,874 filed Mar. 4, 1996, now U.S. Pat. No. 5,830,721; which is the national phase of PCT/US95/02126 filed Feb. 17, 1995; and a CIP of Ser. No. 08/425,684 filed Apr. 18, 1995, now U.S. Pat. No. 5,834,252.

INT-CL: [07] C12 N 15/00, C12 Q 1/68, C07 H 21/02, C07 H 21/04

US-CL-ISSUED: 435/440; 435/6, 536/23.1, 536/24.3, 935/76, 935/77, 935/78

US-CL-CURRENT: 435/440; 435/6, 536/23.1, 536/24.3

FIELD-OF-SEARCH: 435/440, 435/6, 536/23.1, 536/24.3, 935/76, 935/77, 935/78

PRIOR-ART-DISCLOSED:

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Ho, S. et al., "Site-directed Mutagenesis by Overlap Extension Using the Polymerase Chain Reaction," *Gene* 77:51-59 (1989).

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Stemmer, W.P.C., "Searching Sequence Space: Using Recombination to Search More Efficiently and Thoroughly Instead of Making Bigger Combinatorial Libraries," Bio/Technology 13:549-553 (1995).

Stemmer, W.P.C., "Rapid Evolution of a Protein In Vitro by DNA Shuffling," Nature 370:389-391 (1994).

Stemmer, W.P.C., "The Evolution of Molecular Computation," Science 270:1510 (1995).

Stemmer, W.P.C., "DNA Shuffling by Random Fragmentation and Reassembly: In Vitro Recombination for Molecular Evolution," Proc. Natl. Acad. Sci. U.S.A. 91:10747-10751 (1994).

ART-UNIT: 165

PRIMARY-EXAMINER: Whisenant; Ethan

ABSTRACT:

The present invention is generally directed to the evolution of new metabolic pathways and the enhancement of bioprocessing through a process herein termed recursive sequence recombination. Recursive sequence recombination entails performing iterative cycles of recombination and screening or selection to "evolve" individual genes, whole plasmids or viruses, multigene clusters, or even whole genomes. Such techniques do not require the extensive analysis and computation required by conventional methods for metabolic engineering.

134 Claims, 1 Drawing figures

WEST[Generate Collection](#)[Print](#)**Search Results - Record(s) 1 through 10 of 20 returned.**☐ 1. Document ID: US 6488850 B2

L2: Entry 1 of 20

File: USPT

Dec 3, 2002

US-PAT-NO: 6488850

DOCUMENT-IDENTIFIER: US 6488850 B2

TITLE: Method and apparatus for anaerobically degrading pollutants with alkanes

DATE-ISSUED: December 3, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Perriello; Felix Anthony	Norwood	MA		

US-CL-CURRENT: [210/605](#); [210/170](#), [210/220](#), [210/611](#), [210/747](#), [210/908](#), [435/262](#), [435/262.5](#)

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	Claims	KWIC
Draw Desc	Image										

☐ 2. Document ID: US 6245235 B1

L2: Entry 2 of 20

File: USPT

Jun 12, 2001

US-PAT-NO: 6245235

DOCUMENT-IDENTIFIER: US 6245235 B1

TITLE: System and method of in-situ bioremediation with butane-utilizing bacteria

DATE-ISSUED: June 12, 2001

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Perriello; Felix Anthony	Norwood	MA	02062	

US-CL-CURRENT: [210/611](#); [210/620](#), [210/747](#), [210/908](#), [210/909](#), [435/262.5](#)

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	Claims	KWIC
Draw Desc	Image										

☐ 3. Document ID: US 6241779 B1

L2: Entry 3 of 20

File: USPT

Jun 5, 2001

US-PAT-NO: 6241779

DOCUMENT-IDENTIFIER: US 6241779 B1

TITLE: Metal ligand containing bleaching compositions

DATE-ISSUED: June 5, 2001

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Collins; Terrence J.	Pittsburgh	PA		
Horwitz; Colin P.	Pittsburgh	PA		

US-CL-CURRENT: 8/111; 252/186.33, 252/186.39, 252/186.43, 510/311, 8/107, 8/108.1

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	KWIC
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☐ 4. Document ID: US 6210579 B1

L2: Entry 4 of 20

File: USPT

Apr 3, 2001

US-PAT-NO: 6210579

DOCUMENT-IDENTIFIER: US 6210579 B1

TITLE: Bioremediation of pollutants with butane-utilizing bacteria

DATE-ISSUED: April 3, 2001

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Perriello; Felix Anthony	West Roxbury	MA		

US-CL-CURRENT: 210/611; 210/620, 210/747, 210/908, 210/909, 435/262.5

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	KWIC
Draw Desc	Image									

☐ 5. Document ID: US 6136223 A

L2: Entry 5 of 20

File: USPT

Oct 24, 2000

US-PAT-NO: 6136223

DOCUMENT-IDENTIFIER: US 6136223 A

TITLE: Metal ligand containing bleaching compositions

DATE-ISSUED: October 24, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Collins; Terrence J.	Pittsburgh	PA		
Horwitz; Colin P.	Pittsburgh	PA		

US-CL-CURRENT: 252/186.33; 252/186.39, 252/186.43, 510/311, 540/460, 540/465

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments
Draw Desc	Image								

KMC

☐ 6. Document ID: US 6110372 A

L2: Entry 6 of 20

File: USPT

Aug 29, 2000

US-PAT-NO: 6110372

DOCUMENT-IDENTIFIER: US 6110372 A

TITLE: Bioremediation of petroleum pollutants with alkane-utilizing bacteria

DATE-ISSUED: August 29, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Perriello; Felix Anthony	Norwood	MA	02062	

US-CL-CURRENT: 210/611; 210/620, 210/747, 210/908, 210/909, 435/262, 435/262.5

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments
Draw Desc	Image								

KMC

☐ 7. Document ID: US 6107528 A

L2: Entry 7 of 20

File: USPT

Aug 22, 2000

US-PAT-NO: 6107528

DOCUMENT-IDENTIFIER: US 6107528 A

TITLE: Iron complexes for bleach activation and stereospecific oxidation

DATE-ISSUED: August 22, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Que, Jr.; Lawrence	Roseville	MN		
Kim; Cheal	Minneapolis	MN		
Kim; Jinheung	Chapel Hill	NC		
Zang; Yan	Minneapolis	MN		

US-CL-CURRENT: 568/832; 252/186.38, 252/186.39, 252/186.4, 252/186.41, 252/186.42, 510/311, 510/376, 556/138, 8/111

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments
Draw Desc	Image								

KMC

☐ 8. Document ID: US 6100394 A

L2: Entry 8 of 20

File: USPT

Aug 8, 2000

US-PAT-NO: 6100394

DOCUMENT-IDENTIFIER: US 6100394 A

TITLE: Long-lived homogenous oxidation catalysts

DATE-ISSUED: August 8, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Collins; Terrence J.	Pittsburgh	PA		
Gordon-Wylie; Scott W.	Pittsburgh	PA		

US-CL-CURRENT: 540/467; 502/150, 540/450, 540/451, 540/452, 540/453, 540/465, 540/480, 540/482, 540/483

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments
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KWIC

☐ 9. Document ID: US 6099586 A

L2: Entry 9 of 20

File: USPT

Aug 8, 2000

US-PAT-NO: 6099586

DOCUMENT-IDENTIFIER: US 6099586 A

TITLE: Metal ligand containing bleaching compositions

DATE-ISSUED: August 8, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Collins; Terrence J.	Pittsburgh	PA		
Horwitz; Colin P.	Pittsburgh	PA		

US-CL-CURRENT: 8/111; 252/186.33, 252/186.39, 252/186.43, 510/311, 8/108.1

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments
Draw Desc	Image								

KWIC

☐ 10. Document ID: US 6054580 A

L2: Entry 10 of 20

File: USPT

Apr 25, 2000

US-PAT-NO: 6054580

DOCUMENT-IDENTIFIER: US 6054580 A

TITLE: Long-lived homogenous amide containing macrocyclic compounds

DATE-ISSUED: April 25, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Collins; Terrence J.	Pittsburgh	PA		
Gordon-Wylie; Scott W.	Burlington	VT		
Horwitz; Colin P.	Pittsburgh	PA		

US-CL-CURRENT: 540/460; 540/465, 540/480, 540/482, 540/483

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	KWIC
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Terms	Documents
L1 and dichlorobenzene	20

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WEST[Generate Collection](#)[Print](#)**Search Results - Record(s) 1 through 2 of 2 returned.**☐ 1. Document ID: US 6117661 A

L9: Entry 1 of 2

File: USPT

Sep 12, 2000

US-PAT-NO: 6117661

DOCUMENT-IDENTIFIER: US 6117661 A

TITLE: Mutant mono-oxygenase cytochrome P450cam

DATE-ISSUED: September 12, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Wong; Luet-Lok	Oxford			GB
Flitsch; Sabine Lahja	Edinburgh			GB
Nickerson; Darren Paul	Oxford			GB
Hart; Alwyn James	Loughborough			GB

US-CL-CURRENT: 435/189; 435/252.3, 435/320.1, 435/471, 435/69.1, 536/23.2

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	KWIC
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☐ 2. Document ID: US 6100074 A

L9: Entry 2 of 2

File: USPT

Aug 8, 2000

US-PAT-NO: 6100074

DOCUMENT-IDENTIFIER: US 6100074 A

TITLE: Mutant mono-oxygenase cytochrome P-450 .sub.cam

DATE-ISSUED: August 8, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Flitsch; Sabine Lahja	Edinburgh			GB
Nickerson; Darren Paul	Oxford			GB
Wong; Luet-Lok	Oxford			GB

US-CL-CURRENT: 435/189; 435/132, 435/69.1, 530/402

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	KWIC
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Terms	Documents
mutant mono-oxygenase	2

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L9: Entry 1 of 2

File: USPT

Sep 12, 2000

US-PAT-NO: 6117661

DOCUMENT-IDENTIFIER: US 6117661 A

TITLE: Mutant mono-oxygenase cytochrome P450cam

DATE-ISSUED: September 12, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Wong; Luet-Lok	Oxford			GB
Flitsch; Sabine Lahja	Edinburgh			GB
Nickerson; Darren Paul	Oxford			GB
Hart; Alwyn James	Loughborough			GB

US-CL-CURRENT: 435/189; 435/252.3, 435/320.1, 435/471, 435/69.1, 536/23.2

CLAIMS:

What is claimed is:

1. A mutant mono-oxygenase cytochrome P450cam comprising either a deletion of the cysteine at amino acid position 334 or a substitution of another amino acid for the cysteine at amino acid position 334.
2. A mutant mono-oxygenase cytochrome P450cam according to claim 1 wherein an amino acid other than cysteine is substituted at amino acid position 334.
3. A mutant mono-oxygenase cytochrome P450cam according to claim 2 further comprising the substitution of an amino acid other than tyrosine at amino acid position 96.
4. A mutant mono-oxygenase cytochrome P.sup.450 cam according to claim 2 further comprising one or more amino acid substitutions at amino acid positions selected from the group consisting of 87, 98, 101, 185, 193, 244, 247, 295, 297, 395, and 396.
5. A mutant mono-oxygenase cytochrome P450cam according to claim 1 wherein the cysteine is deleted at amino acid position 334.
6. A mutant mono-oxygenase cytochrome P450cam according to claim 5 further comprising the substitution of an amino acid other than tyrosine at amino acid position 96.
7. A mutant mono-oxygenase cytochrome P450cam according to claim 5 further comprising one or more amino acid substitutions at amino acid positions selected from the group consisting of 87, 98, 101, 185, 193, 244., 247, 295, 297, 395, and 396.
8. A mutant mono-oxygenase cytochrome P450cam according to claim 1 further comprising the substitution of an amino acid other than tyrosine at amino acid

position 96.

9. A mutant mono-oxygenase cytochrome P450cam according to claim 8 wherein the substituent amino acid for position 96 is selected from the group consisting of alanine, arginine, asparagine, aspartic acid, glutamic acid, glutamine, glycine, histidine, isoleucine, leucine, lysine, methionine, proline, serine, threonine, tryptophan, tyrosine and valine.

10. A mutant mono-oxygenase cytochrome P450cam according to claim 8 further comprising one or more amino acid substitutions at amino acid positions selected from the group consisting of 87, 98, 101, 185, 193, 244, 247, 295, 297, 395, and 396.

11. A mutant mono-oxygenase cytochrome P450cam according to claim 1 wherein the substituent amino acid is selected from the group consisting of alanine, arginine, asparagine, aspartic acid, glutamic acid, glutamine, glycine, histidine, isoleucine, leucine, lysine, methionine, proline, serine, threonine, tryptophan, tyrosine and valine.

12. A mutant mono-oxygenase cytochrome P450cam according to claim 11 further comprising one or more amino acid substitutions at amino acid positions selected from the group consisting of 87, 98, 101, 185, 193, 244, 247, 295, 297, 395, and 396.

13. A mutant mono-oxygenase cytochrome P450cam according to claim 1 further comprising one or more amino acid substitutions at amino acid positions selected from the group consisting of 87, 98, 101, 185, 193, 244, 247, 295, 297, 395, and 396.

WEST**End of Result Set**

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L9: Entry 2 of 2

File: USPT

Aug 8, 2000

US-PAT-NO: 6100074

DOCUMENT-IDENTIFIER: US 6100074 A

TITLE: Mutant mono-oxygenase cytochrome P-450 .sub.cam

DATE-ISSUED: August 8, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Flitsch; Sabine Lahja	Edinburgh			GB
Nickerson; Darren Paul	Oxford			GB
Wong; Luet-Lok	Oxford			GB

US-CL-CURRENT: 435/189; 435/132, 435/69.1, 530/402

CLAIMS:

We claim:

1. A mutant mono-oxygenase cytochrome P-450.sub.cam wherein the tyrosine residue at position 96 is replaced by the residue of a small hydrophobic amino acid.
2. The mutant of claim 1, wherein said mutant catalyzes the oxidation of a compound selected from the group consisting of a polycyclic aromatic hydrocarbon, a linear or branched alkane, a biphenyl compound and a halogenated hydrocarbon.
3. The mutant of claim 1, wherein the amino acid is selected from the group consisting of alanine, glycine, isoleucine, leucine, and valine.
4. The mutant of claim 1, wherein an amino acid residue at one or more of the positions 87, 98, 185, 244, 247, 295 or 297 is independently replaced by another amino acid residue.
5. The mutant of claim 2, wherein the amino acid is selected from the group consisting of alanine, glycine, isoleucine, leucine, and valine.
6. The mutant of claim 2, wherein an amino acid residue at one or more of the positions 87, 98, 185, 244, 247, 295 or 297 is independently replaced by another amino acid residue.
7. The mutant of claim 3, wherein an amino acid residue at one or more of the positions 87, 98, 185, 244, 247, 295 or 297 is independently replaced by another amino acid residue.
8. A method of oxidizing a compound selected from the group consisting of a polycyclic aromatic hydrocarbon, a linear or branched alkane, a biphenyl compound or a halogenated variant thereof and a halogenated hydrocarbon, comprising the step of contacting said compound under oxidizing conditions with

mono-oxygenase cytochrome P-450.sub.cam wherein the tyrosine residue at position 96 is replaced by a small hydrophobic amino acid residue.

9. The method of claim 8, wherein the amino acid is selected from the group consisting of alanine, glycine, isoleucine, leucine, and valine.

10. The method of claim 8, wherein an amino acid residue at one or more of the positions 87, 98, 185, 244, 247, 295 or 297 is independently replaced by another amino acid residue.

11. The method of claim 9, wherein an amino acid residue at one or more of the positions 87, 98, 185, 244, 247, 295 or 297 is independently replaced by another amino acid residue.

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L11: Entry 11 of 86

File: USPT

May 28, 2002

US-PAT-NO: 6395299

DOCUMENT-IDENTIFIER: US 6395299 B1

TITLE: Matrices for drug delivery and methods for making and using the same

DATE-ISSUED: May 28, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Babich; John W.	Scituate	MA		
Zubietta; Jon	Syracuse	NY		
Bonavia; Grant	Kensington	MD		

US-CL-CURRENT: 424/484

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	KWC
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☐ **12. Document ID: US 6388171 B1**

L11: Entry 12 of 86

File: USPT

May 14, 2002

US-PAT-NO: 6388171

DOCUMENT-IDENTIFIER: US 6388171 B1

TITLE: Compositions and methods for fumonisin detoxification

DATE-ISSUED: May 14, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Duvick; Jon	Des Moines	IA		
Maddox; Joyce	Des Moines	IA		
Gilliam; Jacob	Norwalk	IA		
Folkerts; Otto	Guilford	CT		
Crasta; Oswald R.	Branford	CT		

US-CL-CURRENT: 800/279; 435/320.1, 435/418, 435/419, 435/468, 435/69.1, 536/23.2, 536/23.7, 536/24.1, 800/278, 800/287, 800/288, 800/306, 800/312, 800/314, 800/317.4, 800/320, 800/320.1, 800/320.2, 800/320.3, 800/322

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	KWC
Draw. Desc	Image									

☐ 13. Document ID: US 6380145 B1

L11: Entry 13 of 86

File: USPT

Apr 30, 2002

US-PAT-NO: 6380145

DOCUMENT-IDENTIFIER: US 6380145 B1

TITLE: Cleaning compositions comprising a specific oxygenase

DATE-ISSUED: April 30, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Herbots; Ivan Maurice Alfons Jan	B-1853 Strombeek-Bever			BE
Barnabas; Mary Vijayarani	Cincinnati	OH	45061	
Bettiol; Jean-Luc Philippe	B-1853 Strombeek-Bever			BE
Busch; Alfred	B-1853 Strombeek-Bever			BE

US-CL-CURRENT: 510/392; 510/114, 510/226, 510/300, 510/305, 510/306, 510/320, 510/374,
510/530

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments
Drawn Desc	Image								

KWC

☐ 14. Document ID: US 6365377 B1

L11: Entry 14 of 86

File: USPT

Apr 2, 2002

US-PAT-NO: 6365377

DOCUMENT-IDENTIFIER: US 6365377 B1

TITLE: Recombination of insertion modified nucleic acids

DATE-ISSUED: April 2, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Patten; Phillip A.	Mountain View	CA		
Heinrichs; Volker	Mountain View	CA		
Stemmer; Willem P. C.	Los Gatos	CA		

US-CL-CURRENT: 435/91.1; 435/455, 435/463, 435/6, 436/94

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments
Drawn Desc	Image								

KWC

☐ 15. Document ID: US 6365376 B1

L11: Entry 15 of 86

File: USPT

Apr 2, 2002

US-PAT-NO: 6365376

DOCUMENT-IDENTIFIER: US 6365376 B1

TITLE: Genes and enzymes for the production of adipic acid intermediates

DATE-ISSUED: April 2, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Brzostowicz; Patricia C.	West Chester	PA		
Rouviere; Pierre E.	Wilmington	DE		

US-CL-CURRENT: 435/91.1, 435/252.3, 435/252.31, 435/252.32, 435/252.33, 435/252.35,
435/254.11, 435/254.2, 435/320.1, 435/91.2, 536/23.2, 536/23.7

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments
Draw Desc	Image								

KWIC

☐ 16. Document ID: US 6310271 B1

L11: Entry 16 of 86

File: USPT

Oct 30, 2001

US-PAT-NO: 6310271

DOCUMENT-IDENTIFIER: US 6310271 B1

TITLE: Polynucleotides encoding choline monooxygenase and plants transformed therewith

DATE-ISSUED: October 30, 2001

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Hanson; Andrew D.	Gainesville	FL		
Rathinasabapathi; Bala	Gainesville	FL		
Burnet; Michael	Les Hameaux			FR

US-CL-CURRENT: 800/278, 435/410, 435/419, 435/468, 435/69.1, 536/23.1, 536/23.6,
800/285, 800/290, 800/295, 800/306, 800/312, 800/314, 800/317.2, 800/317.3, 800/317.4,
800/320, 800/320.1, 800/320.2, 800/320.3, 800/322

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments
Draw Desc	Image								

KWIC

☐ 17. Document ID: US 6309883 B1

L11: Entry 17 of 86

File: USPT

Oct 30, 2001

US-PAT-NO: 6309883

DOCUMENT-IDENTIFIER: US 6309883 B1

TITLE: Methods and compositions for cellular and metabolic engineering

DATE-ISSUED: October 30, 2001

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Minshull; Jeremy	San Francisco	CA		
Stemmer; Willem P. C.	Los Gatos	CA		

US-CL-CURRENT: 435/440; 435/6, 536/23.1, 536/24.3

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments
Draw Desc	Image								

KWC

☐ 18. Document ID: US 6300544 B1

L11: Entry 18 of 86

File: USPT

Oct 9, 2001

US-PAT-NO: 6300544

DOCUMENT-IDENTIFIER: US 6300544 B1

TITLE: Cytochrome P450 monooxygenases

DATE-ISSUED: October 9, 2001

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Halkier; Barbara Ann	Copenhagen K.			DK
Bak; Soren	Copenhagen N.			DK
Kahn; Rachel Alice	Copenhagen K.			DK
Moller; Birger Lindberg	Bronshoj			DK

US-CL-CURRENT: 800/279; 435/183, 435/252.3, 435/320.1, 435/419, 435/6, 530/350, 530/370, 536/23.6, 536/24.1, 800/301

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments
Draw Desc	Image								

KWC

☐ 19. Document ID: US 6268552 B1

L11: Entry 19 of 86

File: USPT

Jul 31, 2001

US-PAT-NO: 6268552

DOCUMENT-IDENTIFIER: US 6268552 B1

TITLE: Transgenic seedless fruit comprising AGL or GH3 promoter operably linked to isopentenyl transferase or tryptophan monooxygenase coding DNA

DATE-ISSUED: July 31, 2001

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Li; Yi	Mansfield Center	KS		

US-CL-CURRENT: 800/317.4; 435/320.1, 800/278, 800/284, 800/298, 800/307, 800/308

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments
Draw Desc	Image								

KWC

☐ 20. Document ID: US 6255067 B1

L11: Entry 20 of 86

File: USPT

Jul 3, 2001

US-PAT-NO: 6255067

DOCUMENT-IDENTIFIER: US 6255067 B1

TITLE: cDNA encoding peptidyl-glycine alpha-amidating monooxygenase (PAM)

DATE-ISSUED: July 3, 2001

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Keutmann; Henry T.	Concord	MA		
Schofield; Peter	Heidelberg			DE
Rodriguez; Henry	Belmont	CA		
Eipper; Betty	Baltimore	MD		
Mains; Richard	Baltimore	MD		

US-CL-CURRENT: 435/69.1; 435/252.3, 435/254.2, 435/320.1, 435/325, 435/349, 530/350,
536/23.5

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	KWIC
Drawn Desc	Image									

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Terms	Documents
monooxygenase.clm.	86

Display Format: [CIT](#) [Change Format](#)[Previous Page](#)[Next Page](#)

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L15: Entry 2 of 20

File: USPT

Oct 29, 2002

US-PAT-NO: 6472191

DOCUMENT-IDENTIFIER: US 6472191 B1

TITLE: DNA FRAGMENT CARRYING TOLUENE MONOOXYGENASE GENE, RECOMBINANT PLASMID, TRANSFORMED MICROORGANISM, METHOD FOR DEGRADING CHLORINATED ALIPHATIC HYDROCARBON COMPOUNDS AND AROMATIC COMPOUNDS, AND METHOD FOR ENVIRONMENTAL REMEDIATION

DATE-ISSUED: October 29, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Yano; Tetsuya	Atsugi			JP
Nomoto; Tsuyoshi	Komae			JP
Imamura; Takeshi	Chigasaki			JP

US-CL-CURRENT: 435/189; 435/252.3, 435/262.5, 435/320.1, 536/23.2

CLAIMS:

What is claimed is:

1. An isolated DNA fragment of about 5.3 Kb containing a toluene monooxygenase gene, having 3 BamHI, 1 ClaI, 1 EcoRI, 3 KpnI, 2 NcoI, 2 NspV, 2 ScaI, 2 SmaI, 2 SphI, 1 StuI, 0 DraI, 0 EcoRV, 0 HindIII, 0 HpaI, 0 NdeI, 0 PvuII, 0 ScaI, 0 Sse8387I, 0 XbaI, 0 XhoI restriction sites, and having a restriction map of: ##STR3##
2. An isolated DNA fragment having a nucleotide sequence of SEQ ID NO: 1 in the Sequence Listing.
3. A DNA fragment encoding a protein having a toluene monooxygenase activity and being hybridizable to a nucleotide sequence from 200 . . . 4799 of SEQ ID NO: 1 or a complementary sequence thereof under stringent hybridization conditions.
4. A recombinant DNA comprising a vector which can replicate or can be maintained in a host and a DNA fragment according to any one of claims 1 to 3.
5. The recombinant DNA fragment according to claim 4, wherein the vector can be maintained or replicate in a bacterium.
6. A transformant obtainable by introducing into a host microorganism a recombinant DNA comprising the DNA fragment according to any one of claims 1 to 3 ligated to a vector which can replicate or be maintained in the host.
7. A method for producing a toluene monooxygenase, comprising: introducing into a host microorganism a recombinant DNA which comprises a vector which can replicate or can be maintained in the host microorganism and a DNA fragment according to any one of claims 1 to 3 to form a transformant which produces a toluene monooxygenase encoded by said DNA fragment.

8. A method for remedying an environment polluted with a pollutant being at least either of a halogenated aliphatic hydrocarbon compound or an aromatic compound, comprising a step of degrading the pollutant by bringing a transformant into contact with the pollutant, wherein the transformant is obtainable by introducing into a host microorganism a recombinant DNA constituted by ligating a vector which enables retention and replication in the host and a DNA fragment according to any one of claims 1 to 3.
9. The remediation method according to claim 8, wherein the environment is soil.
10. The remediation method according to claim 9 comprising the steps of: introducing an aqueous medium containing the transformant into the polluted soil; and supplying nutrients and/or oxygen for proliferation of the transformant in the polluted soil.
11. The remediation method according to claim 10 wherein the transformant is introduced in the soil by applying pressure through an injection well provided in the polluted soil.
12. The remediation method according to claim 9 wherein the polluted soil is introduced in a liquid phase containing the transformant.
13. The remediation method according to claim 9 wherein the polluted soil is brought into contact with a carrier holding the transformant.
14. The remediation method according to claim 8 wherein the environment is air.
15. The remediation method according to claim 14 wherein the polluted air is introduced into a liquid phase containing the transformant.
16. The remediation method according to claim 14 wherein the polluted air is brought into contact with a carrier holding the transformant.
17. The remediation method according to claim 16 wherein contact is carried out by placing the carrier holding the transformant in a container, introducing polluted air from one side of the container, and discharging cleaned air from another side.
18. The remediation method according to claim 8 wherein the halogenated aliphatic hydrocarbon compound is either trichloroethylene (TCE) or dichloroethylene (DCE).
19. The remediation method according to claim 8 wherein the aromatic compound is at least one of toluene, benzene, phenol, and cresol.

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L15: Entry 9 of 20

File: USPT

Sep 28, 1999

US-PAT-NO: 5958757

DOCUMENT-IDENTIFIER: US 5958757 A

TITLE: Biological conversion of organic compounds

DATE-ISSUED: September 28, 1999

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Steffan; Robert Jon	Newtown	PA		
McClay; Kevin Rock	Morrisville	PA		

US-CL-CURRENT: 435/262.5; 435/170, 435/264, 435/289.1, 435/874, 570/220, 588/248

CLAIMS:

We claim:

1. A method for oxidizing saturated aliphatic halocarbons comprising contacting said halocarbons with an aromatic oxygenase capable of oxidizing said halocarbons, which is produced by a microorganism wherein said saturated halocarbon is selected from the group consisting of chloroform, bromoform, 1,2-dichloroethane, 1,2 dibromoethane, monochloroethane, and monobromoethane.
2. The method of claim 1 wherein said saturated aliphatic halocarbon is chloroform.
3. The method of claim 1 which further comprises providing a co-substrate to support degradation of said halocarbon by said microorganism.
4. The method of claim 3 wherein said co-substrate is selected from the group consisting of toluene, phenol, benzene, ethylbenzene, and xylene.
5. The method of claim 1 wherein said saturated halocarbon is contacted with said microorganism in water.
6. The method of claim 1 wherein said saturated halocarbon is contacted with said microorganism in soil.
7. The method of claim 1 wherein said saturated halocarbon is contacted with said microorganism in vapor phase.
8. The method of claim 1 wherein said saturated halocarbon is contacted with said aromatic oxygenase-producing microorganism within a bioreactor.
9. The method of claim 8 wherein said bioreactor is a fixed film bioreactor.
10. The method of claim 8 wherein said bioreactor is a suspended growth bioreactor.
11. The method of claim 1 wherein said saturated halocarbon is contacted with

said aromatic oxygenase-producing bacteria in situ.

12. The method of claim 11 wherein said saturated halocarbon is present in soil or sludge.

13. The method of claim 11 wherein said saturated halocarbon is present in groundwater.

14. The method of claim 1 wherein said aromatic oxygenase is a toluene monooxygenase.

15. The method of claim 1 wherein said aromatic oxygenase-producing microorganism is selected from the group consisting of *Pseudomonas mendocina* KR1, ATCC 55706; Strain ENVPC5; and Strain ENVBF1 ATCC 55819.

16. The method of claim 1 wherein said aromatic oxygenase-producing microorganism is a recombinant microorganism consisting of a host microorganism containing cloned aromatic oxygenase genes.

17. A method for oxidizing saturated aliphatic halocarbons comprising contacting said halocarbons with an aromatic oxygenase capable of oxidizing said halocarbons, which is produced by an aerobic bacteria wherein said saturated halocarbon is selected from the group consisting of chloroform, bromoform, 1,2-dichloroethane, 1,2 dibromoethane, monochloroethane, and monobromoethane.

18. A method for oxidizing saturated aliphatic halocarbons comprising contacting said halocarbons with a toluene-4-monooxygenase which is produced by an aerobic bacteria.

WEST Search History

DATE: Tuesday, January 14, 2003

<u>Set Name</u> side by side	<u>Query</u>	<u>Hit Count</u>	<u>Set Name</u> result set
	<i>DB=USPT; PLUR=YES; OP=ADJ</i>		
L17	L13 and aromatic sibstrate	0	L17
L16	L13 and halo aromatic sibstrate	0	L16
L15	l11 and oxidation and benzene	20	L15
L14	l11 and oxidation and dichlorobenzene	0	L14
L13	l11 and oxidation	51	L13
L12	mutant monooxygenase.clm.	0	L12
L11	monooxygenase.clm.	86	L11
L10	mono-oxygenase.clm.	9	L10
L9	mutant mono-oxygenase	2	L9
L8	oxidizing dichlorophenyl	0	L8
L7	oxidizing pentachlorophenyl	0	L7
L6	L1 and pentachlorophenyl	1	L6
L5	L1 and dichlorophenyl	7	L5
L4	L2 and pentachlorobiphenyl	0	L4
L3	L2 and dichlorobiphenyl	0	L3
L2	L1 and dichlorobenzene	20	L2
L1	monooxygenase	653	L1

END OF SEARCH HISTORY

WEST Search History

DATE: Tuesday, January 14, 2003

<u>Set Name</u>	<u>Query</u>	<u>Hit Count</u>	<u>Set Name</u>
side by side			result set
	<i>DB=USPT; PLUR=YES; OP=ADJ</i>		
L21	L20 and p-450	3	L21
L20	oxidizing and monooxygenase.clm.	33	L20
L19	oxidizing and monooxygenase	193	L19
L18	method for oxidizing and monooxygenase	0	L18
L17	L13 and aromatic sibstrate	0	L17
L16	L13 and halo aromatic sibstrate	0	L16
L15	l11 and oxidation and benzene	20	L15
L14	l11 and oxidation and dichlorobenzene	0	L14
L13	l11 and oxidation	51	L13
L12	mutant monooxygenase.clm.	0	L12
L11	monooxygenase.clm.	86	L11
L10	mono-oxygenase.clm.	9	L10
L9	mutant mono-oxygenase	2	L9
L8	oxidizing dichlorophenyl	0	L8
L7	oxidizing pentachlorophenyl	0	L7
L6	L1 and pentachlorophenyl	1	L6
L5	L1 and dichlorophenyl	7	L5
L4	L2 and pentachlorobiphenyl	0	L4
L3	L2 and dichlorobiphenyl	0	L3
L2	L1 and dichlorobenzene	20	L2
L1	monooxygenase	653	L1

END OF SEARCH HISTORY

WEST**End of Result Set**

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L5: Entry 1 of 1

File: DWPI

May 7, 1997

DERWENT-ACC-NO: 1997-229326

DERWENT-WEEK: 200046

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TITLE: Mutant of the mono-oxygenase cytochrome P-450cam - useful in catalysis of oxidation of a wide range of organic substrates, such as lindane.

INVENTOR: FLITSCH, S L; HART, A J ; NICKERSON, D P ; WONG, L

PRIORITY-DATA: 1995WO-GB02588 (November 2, 1995), 1995GB-0022407 (November 1, 1995)

PATENT-FAMILY:

PUB-NO	PUB-DATE	LANGUAGE	PAGES	MAIN-IPC
GB 2306485 A	May 7, 1997		040	C12N009/02
US 6117661 A	September 12, 2000		000	C12N009/02
WO 9716553 A1	May 9, 1997		000	C12N015/53
AU 9673236 A	May 22, 1997		000	C12N015/53
GB 2306485 B	December 9, 1998		000	C12N009/02
CZ 9801273 A3	January 13, 1999		000	C12N015/53
EP 906431 A1	April 7, 1999	E	000	C12N015/53
SK 9800555 A3	April 13, 1999		000	C12N015/53
CN 1212015 A	March 24, 1999		000	C12N015/53
NZ 320497 A	September 29, 1999		000	C12N015/53
AU 716583 B	March 2, 2000		000	C12N015/53
JP 2000508163 W	July 4, 2000		039	C12N009/02

INT-CL (IPC): C12 N 9/02; C12 N 15/00; C12 N 15/09; C12 N 15/53; C12 N 15/78; C12 P 7/02; C12 P 7/22; C12 R 1:19; C12 R 1:19

ABSTRACTED-PUB-NO: GB 2306485A

BASIC-ABSTRACT:

Mutant of the mono-oxygenase cytochrome P-450cam, in which the cysteine residue at position 334 is removed, is new.

USE- Mono-oxygenase cytochrome P-450cam from *P. putida* catalyses the regio- and stereoselective hydroxylation of camphor to 5-exo-hydroxycamphor. The mutant enzyme may be used to catalyse the oxidation of a relatively wide range of organic substrates (such as lindane), whether or not these are substrates for the wild-type protein.

ADVANTAGE- A C334A mutant of P-450cam did not show any evidence of aggregation even at mM concentrations at room temperature over a period of three days. This property is expected to improve protein handling, storage and increased catalyst lifetime.

ABSTRACTED-PUB-NO:

GB 2306485B

EQUIVALENT-ABSTRACTS:

Mutant of the mono-oxygenase cytochrome P-450cam, in which the cysteine residue at position 334 is removed, is new.

USE- Mono-oxygenase cytochrome P-450cam from *P. putida* catalyses the regio- and stereoselective hydroxylation of camphor to 5-exo-hydroxycamphor. The mutant enzyme may be used to catalyse the oxidation of a relatively wide range of organic substrates (such as lindane), whether or not these are substrates for the wild-type protein.

ADVANTAGE- A C334A mutant of P-450cam did not show any evidence of aggregation even at mM concentrations at room temperature over a period of three days. This property is expected to improve protein handling, storage and increased catalyst lifetime.

US 6117661A

Mutant of the mono-oxygenase cytochrome P-450cam, in which the cysteine residue at position 334 is removed, is new.

USE- Mono-oxygenase cytochrome P-450cam from *P. putida* catalyses the regio- and stereoselective hydroxylation of camphor to 5-exo-hydroxycamphor. The mutant enzyme may be used to catalyse the oxidation of a relatively wide range of organic substrates (such as lindane), whether or not these are substrates for the wild-type protein.

ADVANTAGE- A C334A mutant of P-450cam did not show any evidence of aggregation even at mM concentrations at room temperature over a period of three days. This property is expected to improve protein handling, storage and increased catalyst lifetime.

ABSTRACTED-PUB-NO: GB 2306485A

EQUIVALENT-ABSTRACTS: GB 2306485B Mutant of the mono-oxygenase cytochrome P-450cam, in which the cysteine residue at position 334 is removed, is new. USE- Mono-oxygenase cytochrome P-450cam from *P. putida* catalyses the regio- and stereoselective hydroxylation of camphor to 5-exo-hydroxycamphor. The mutant enzyme may be used to catalyse the oxidation of a relatively wide range of organic substrates (such as lindane), whether or not these are substrates for the wild-type protein. ADVANTAGE- A C334A mutant of P-450cam did not show any evidence of aggregation even at mM concentrations at room temperature over a period of three days. This property is expected to improve protein handling, storage and increased catalyst lifetime. US 6117661A Mutant of the mono-oxygenase cytochrome P-450cam, in which the cysteine residue at position 334 is removed, is new. USE- Mono-oxygenase cytochrome P-450cam from *P. putida* catalyses the regio- and stereoselective hydroxylation of camphor to 5-exo-hydroxycamphor. The mutant enzyme may be used to catalyse the oxidation of a relatively wide range of organic substrates (such as lindane), whether or not these are substrates for the wild-type protein. ADVANTAGE- A C334A mutant of P-450cam did not show any evidence of aggregation even at mM concentrations at room temperature over a period of three days. This property is expected to improve protein handling, storage and increased catalyst lifetime.

CHOSEN-DRAWING: Dwg.0/2

WEST**End of Result Set**

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L7: Entry 2 of 2

File: DWPI

May 8, 1996

DERWENT-ACC-NO: 1996-211678

DERWENT-WEEK: 200112

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TITLE: New mutant forms of monooxygenase cytochrome P-450 (cam) - with amino acid substitutions at positions 96 and 334, useful for oxidn. of wide range of aliphatic and aromatic, opt. halogenated, hydrocarbon(s)

INVENTOR: FLITSCH, S L; NICKERSON, D P ; WONG, L ; WONG, L L ; NICKERSON, D

PRIORITY-DATA: 1994GB-0022205 (November 3, 1994)

PATENT-FAMILY:

PUB-NO	PUB-DATE	LANGUAGE	PAGES	MAIN-IPC
GB 2294692 A	May 8, 1996		042	C12N009/02
KR 234348 B1	December 15, 1999		000	C12N015/53
WO 9614419 A1	May 17, 1996	E	044	C12N015/53
AU 9538117 A	May 31, 1996		000	C12N015/53
EP 789770 A1	August 20, 1997	E	000	C12N015/53
CZ 9701277 A3	October 15, 1997		000	C12N015/53
SK 9700545 A3	February 4, 1998		000	C12N015/53
JP 10503658 W	April 7, 1998		039	C12N015/09
NZ 294904 A	September 24, 1998		000	C12P007/02
KR 97707288 A	December 1, 1997		000	C12N015/53
GB 2294692 B	January 20, 1999		000	C12N009/02
AU 705736 B	June 3, 1999		000	C12N015/53
RU 2133774 C1	July 27, 1999		000	C12N015/53
US 6100074 A	August 8, 2000		000	C12N009/02

INT-CL (IPC): C07 K 1/00; C12 N 9/02; C12 N 15/09; C12 N 15/53; C12 P 7/00; C12 P 7/02; C12 P 7/22; C12 P 21/06; C12 R 1:19; C12 R 1:19

ABSTRACTED-PUB-NO: GB 2294692A

BASIC-ABSTRACT:

New mutants (I) of mono-oxygenase cytochrome P-450 can have 96 Tyr and/or 334 Cys replaced by any other amino acid (aa) provided: (a) replacement is not by Phe or (b) the mutant is able to oxidise any of polycyclic aromatic hydrocarbons, branched or linear alkanes, diphenyls or biphenyls (including halogenated derivs.) or halogenated hydrocarbons.

USE - (I) are used to oxidise the substrates named above, e.g. to produce starting materials (partic. in homo-chiral form) for organic synthesis.

ADVANTAGE - (I) can oxidise a wide range of substrates (claimed) (alteration of the aromatic binding site pocket reduces specificity for camphor, the natural substrate). Elimination of 334 Cys improves stability and prevents unwanted dimerisation during purification.

ABSTRACTED-PUB-NO:

GB 2294692B

EQUIVALENT-ABSTRACTS:

New mutants (I) of mono-oxygenase cytochrome P-450 can have 96 Tyr and/or 334 Cys replaced by any other amino acid (aa) provided: (a) replacement is not by Phe or (b) the mutant is able to oxidise any of polycyclic aromatic hydrocarbons, branched or linear alkanes, diphenyls or biphenyls (including halogenated derivs.) or halogenated hydrocarbons.

USE - (I) are used to oxidise the substrates named above, e.g. to produce starting materials (partic. in homo-chiral form) for organic synthesis.

ADVANTAGE - (I) can oxidise a wide range of substrates (claimed) (alteration of the aromatic binding site pocket reduces specificity for camphor, the natural substrate). Elimination of 334 Cys improves stability and prevents unwanted dimerisation during purificn.

US 6100074A

New mutants (I) of mono-oxygenase cytochrome P-450 can have 96 Tyr and/or 334 Cys replaced by any other amino acid (aa) provided: (a) replacement is not by Phe or (b) the mutant is able to oxidise any of polycyclic aromatic hydrocarbons, branched or linear alkanes, diphenyls or biphenyls (including halogenated derivs.) or halogenated hydrocarbons.

USE - (I) are used to oxidise the substrates named above, e.g. to produce starting materials (partic. in homo-chiral form) for organic synthesis.

ADVANTAGE - (I) can oxidise a wide range of substrates (claimed) (alteration of the aromatic binding site pocket reduces specificity for camphor, the natural substrate). Elimination of 334 Cys improves stability and prevents unwanted dimerisation during purificn.

ABSTRACTED-PUB-NO: GB 2294692A

EQUIVALENT-ABSTRACTS: GB 2294692B New mutants (I) of mono-oxygenase cytochrome P-450 can have 96 Tyr and/or 334 Cys replaced by any other amino acid (aa) provided: (a) replacement is not by Phe or (b) the mutant is able to oxidise any of polycyclic aromatic hydrocarbons, branched or linear alkanes, diphenyls or biphenyls (including halogenated derivs.) or halogenated hydrocarbons. USE - (I) are used to oxidise the substrates named above, e.g. to produce starting materials (partic. in homo-chiral form) for organic synthesis. ADVANTAGE - (I) can oxidise a wide range of substrates (claimed) (alteration of the aromatic binding site pocket reduces specificity for camphor, the natural substrate). Elimination of 334 Cys improves stability and prevents unwanted dimerisation during purificn. US 6100074A New mutants (I) of mono-oxygenase cytochrome P-450 can have 96 Tyr and/or 334 Cys replaced by any other amino acid (aa) provided: (a) replacement is not by Phe or (b) the mutant is able to oxidise any of polycyclic aromatic hydrocarbons, branched or linear alkanes, diphenyls or biphenyls (including halogenated derivs.) or halogenated hydrocarbons. USE - (I) are used to oxidise the substrates named above, e.g. to produce starting materials (partic. in homo-chiral form) for organic synthesis. ADVANTAGE - (I) can oxidise a wide range of substrates (claimed) (alteration of the aromatic binding site pocket reduces specificity for camphor, the natural substrate). Elimination of 334 Cys improves stability and prevents unwanted dimerisation during purificn.

CHOSEN-DRAWING: Dwg.0/2

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L15: Entry 1 of 2

File: USPT

Sep 12, 2000

US-PAT-NO: 6117661

DOCUMENT-IDENTIFIER: US 6117661 A

TITLE: Mutant mono-oxygenase cytochrome P450cam

DATE-ISSUED: September 12, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Wong; Luet-Lok	Oxford			GB
Flitsch; Sabine Lahja	Edinburgh			GB
Nickerson; Darren Paul	Oxford			GB
Hart; Alwyn James	Loughborough			GB

US-CL-CURRENT: 435/189; 435/252.3, 435/320.1, 435/471, 435/69.1, 536/23.2

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	Claims	KWC
Draw	Desc	Image									

☐ 2. Document ID: US 6100074 A

L15: Entry 2 of 2

File: USPT

Aug 8, 2000

US-PAT-NO: 6100074

DOCUMENT-IDENTIFIER: US 6100074 A

TITLE: Mutant mono-oxygenase cytochrome P-450 .sub.cam

DATE-ISSUED: August 8, 2000

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Flitsch; Sabine Lahja	Edinburgh			GB
Nickerson; Darren Paul	Oxford			GB
Wong; Luet-Lok	Oxford			GB

US-CL-CURRENT: 435/189; 435/132, 435/69.1, 530/402

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	Claims	KWC
Draw	Desc	Image									

WEST Search History

DATE: Tuesday, January 14, 2003

Set Name Query
side by side

Hit Count Set Name
result set

DB=USPT; PLUR=YES; OP=ADJ

L15	mutants same mono-oxygenase cytochrome	2	L15
L14	New mutants same mono-oxygenase cytochrome	0	L14
L13	New mutants near15 mono-oxygenase cytochrome	0	L13
L12	New mutants adj5 mono-oxygenase cytochrome	0	L12
L11	New mutants and mono-oxygenase cytochrome	0	L11
L10	New mutants of mono-oxygenase cytochrome	0	L10
L9	New mutants (I) of mono-oxygenase cytochrome	0	L9
L8	US 6100074A New mutants (I) of mono-oxygenase cytochrome	0	L8

DB=DWPI; PLUR=YES; OP=ADJ

L7	mono-oxygenase cytochrome	2	L7
L6	mutant mono-oxygenase cytochrome	1	L6
L5	mutant mono-oxygenase cytochrome p-450cam	1	L5
L4	wo 96/14419	0	L4

DB=PGPB,JPAB,EPAB,DWPI; PLUR=YES; OP=ADJ

L3	monooxygenase.clm.	20	L3
L2	monooxygenase mutants	1	L2
L1	monooxygenase mutants and oxidation	0	L1

END OF SEARCH HISTORY